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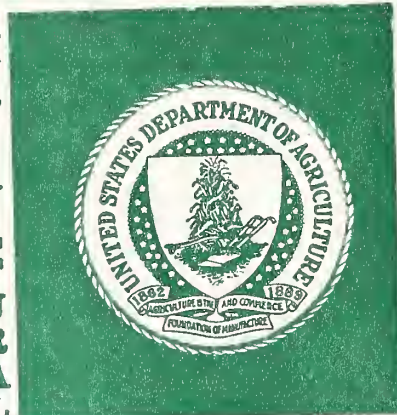
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GRAIN AND FORAGE CROPS RESEARCH
of the
UNITED STATES DEPARTMENT OF AGRICULTURE
and related work of the
State Agricultural Experiment Stations

Section A

This progress report of USDA and cooperative research is primarily a tool for use of scientists and administrators in program coordination, development and evaluation; and for use of advisory committees in program review and development of recommendations for future research programs.

The summaries of progress on USDA and cooperative research include some tentative results that have not been tested sufficiently to justify general release. Such findings, when adequately confirmed will be released promptly through established channels. Because of this, the report is not intended for publication and should not be referred to in literature citations. Copies are distributed only to members of Department staff, advisory committee members and others having a special interest in the development of public agricultural research programs.

This report also includes a list of publications reporting results of USDA and cooperative research issued during the last year. Current agricultural research findings are also published in the monthly USDA publications, Agricultural Research, Agricultural Marketing, and The Farm Index.

UNITED STATES DEPARTMENT OF AGRICULTURE
Washington, D. C.
December 15, 1966

231312
RESEARCH ADVISORY COMMITTEES

The following Research Advisory Committees were established pursuant to Title III of the Research and Marketing Act of 1946:

- | | |
|-----------------------------------|-----------------------------------|
| 1. Farm Resources and Facilities | 8. Cotton |
| 2. Utilization | 9. Grain and Forage Crops |
| 3. Human Nutrition & Consumer Use | 10. Horticultural Crops |
| 4. Marketing | 11. Oilseed, Peanut & Sugar Crops |
| 5. Agricultural Economics | 12. Plant Science & Entomology |
| 6. Forestry | 13. Tobacco |
| 7. Animal and Animal Products | |

The source materials used by the advisory committees include organizational unit progress reports and subject matter progress reports. The latter contain information which was first reported in the organizational reports and has been assembled for use by commodity committees. The number prefixes shown below refer to advisory committees listed above.

ORGANIZATIONAL UNIT PROGRESS REPORTS

Agricultural Research Service (ARS)

- 1 - Agricultural Engineering
- 1 - Soil and Water Conservation
- 2 - Utilization -- Eastern
- 2 - Utilization -- Northern
- 2 - Utilization -- Southern
- 2 - Utilization -- Western
- 3 - Human Nutrition
- 3 - Consumer & Food Economics
- 4 - Market Quality
- 4 - Transportation & Facilities
- 7 - Animal Husbandry
- 7 - Animal Disease & Parasite
- 12 - Crops
- 12 - Entomology

Economic Research Service (ERS)

- 1, 5 - Economic Development
- 4, 5 - Marketing Economics
- 5 - Farm Production Economics
- 5 - Economic & Statistical Analysis
- 5 - Foreign Development & Trade
- 5 - Foreign Regional Analysis
- 5 - Natural Resource Economics
- 6 - Forest Service - Research (FS)
- 4, 5 - Farmer Cooperative Service (FCS)
- 4, 5 - Statistical Reporting Service (SRS)

SUBJECT MATTER PROGRESS REPORTS

- 6 - Forestry (other than Forest Service)
- 7 - Animal-Poultry & Products Research other than
Husbandry, Disease and Parasite
- 8 - Cotton and Cottonseed
- 9 - Grain and Forage Crops
- 10 - Horticultural Crops
- 11 - Oilseed and Peanut
- 11 - Sugar
- 13 - Tobacco

A copy of any of the reports may be requested from W. C. Dachtler, Executive Secretary, Grain and Forage Crops Research Advisory Committee, Research Program Development and Evaluation Staff, U.S. Department of Agriculture, Washington, D. C. 20250.

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INTRODUCTION

This report on grain and forage crops research covers work directly related to the production, processing, distribution and consumption of grain, rice, feed, forage and seed and their products. It does not include extensive cross-commodity work, much of which is basic in character, which contributes to the solution of not only grain and forage crop problems but also to the problems of other commodities. Progress on cross-commodity work is found in the organizations' unit reports of the several divisions.

This report is organized by "Problem Areas" which are shown in the table of contents. For each area there is a statement of (1) the problem, (2) the USDA program, (3) State experiment station programs, (4) a summary of progress during the past year on USDA and cooperative work, (5) a list of publications resulting from USDA and cooperative work and (6) related publications of State agricultural experiment stations.

Research on grain and forage crop problems is supported by (1) Federal funds appropriated to the research agencies of the USDA, (2) Federal and State funds appropriated to the State agricultural experiment stations, and (3) private funds for research carried on in private laboratories or for support of State station and USDA work.

As a step toward implementation of the recommendations for a National Program of Research for Agriculture made jointly by the Association of State Universities and Land Grant Colleges and the USDA, a section has been added to each of the Areas in this report. It comprises a list of the related publications of the State agricultural experiment stations in addition to those heretofore reported covering the results of USDA and cooperative research. In future years, it is anticipated that information will be available to permit reporting of achievements resulting from State research in a format comparable to the present reporting of the USDA and cooperative research.

Research by USDA

Farm research in the Agricultural Research Service comprises investigations on introduction, breeding and genetics, variety evaluation, culture, diseases, nematodes and weed control, insects, and crop handling and harvesting equipment and structures. This research is carried out in the Crops, Entomology, and Agricultural Engineering Research Divisions. It involves approximately 345 scientific man-years.

Nutrition, Consumer and Industrial Use research in the Agricultural Research Service pertains to composition and nutritive value, physiological availability of nutrients and their effects, new and improved methods of preparation, preservation and care in homes, eating establishments and

institutions, new and improved food, feed, and industrial products and the processes related to grains, rice, and forages. It is carried out in the following research divisions: Northern, Southern, and Western Utilization; Human Nutrition; and Consumer and Food Economics. The work involves approximately 190 scientific man-years.

Marketing and Economic research is carried out in four Services. Grain, rice, feed, forage, and seed research in the Agricultural Research Service deals with physical and biological aspects of assembly, packaging, transporting, and storing, and distribution. It is carried out by the Market Quality and Transportation and Facilities Research Divisions. Work in the Economic Research Service deals with marketing costs, margins, and efficiency; market potential; supply and demand; and outlook and situation. Consumer preference studies and research to improve crop estimates are carried out by the Statistical Reporting Service. Research on cooperative marketing is conducted by the Farmer Cooperative Service. The grain and forage research in these Services involves approximately 60 scientific man-years.

Interrelationships Among Department, State and Private Research

A large part of the Department's research is cooperative with State experiment stations. Many Department employees are located at State stations and use laboratories and office space close to or furnished by the State. Cooperative work is jointly planned, frequently with representatives of the producers or industry affected participating. The nature of cooperation varies with each study. It is developed so as to fully utilize the personnel and other resources of the cooperators, which frequently includes resources contributed by the interested producers or industry. There is regular exchange of information between station and Department scientists to assure that the programs compliment each other and to eliminate unnecessary duplication.

Privately supported grain and forage crops research emphasizes the solution of specific production, processing, and marketing problems. Much of it utilizes the results of more basic work done by State stations and Department scientists. For example, private research is devoted to the synthesis and evaluation of chemicals as herbicides, fungicides, and insecticides, the development of new combinations of materials for use as fertilizers, product and process development, improvements in equipment for planting, cultivating, harvesting, handling, processing, drying and storing grains, forages and seeds. Private research in marketing and economics is largely concerned with studies of consumer preferences, market potential, promotion and market development.

The contributions of producers of grain and forage crops and of related industries to the work of State stations and the Department have been an

important factor in the success of public research programs. Producers, processors, and distributors offer land, products, and facilities for the testing of equipment and practices used in the production and distribution of grain, rice, feed, forage, and seed and their products.

Examples of Recent Research Accomplishments

USDA and Cooperating Scientists

Improved Techniques Hasten Progress on Breeding Weevil Resistant Alfalfa. By improving techniques and combining several methods of screening, over 600,000 plants were evaluated in Beltsville, Maryland, laboratories for resistance to the alfalfa weevil. To test the same number of plants in the field would have been prohibitive. Resistance tests can now be conducted throughout the year and with greater precision. Four successive tests are used to isolate resistant plants: 1. Seedlings are grown in growth chambers under high light intensities to reduce hypocotyl elongation and infested with adult weevils at 12 days of age. When all but 1 or 2 percent of the seedlings have been eaten, weevils are removed and the remaining seedlings saved for further testing. 2. Leaf discs from plants selected in Step 1 are tested individually with adult weevils to identify plants which deter adult feeding. 3. Plants selected from Step 2 are tested for larval growth and survival. Individual plants are confined in a plastic cylinder and infested with 10 freshly hatched larvae. Eight days later, larval weight and survival are determined. 4. Plants selected from Steps 2 and 3 are individually tested for oviposition differences. At Raleigh, North Carolina, plants resistant to oviposition preference were isolated.

Use of Plant Introductions in Improving Field and Forage Crops. Significant advances toward the improvement of field and forage crops through the use of exotic germ plasm are reported. The production, quality, and production range of established crops are being improved and increased through the use of desirable exotic germ plasm. Other new crops are being introduced and evaluated. Examples of the use of specific introductions in the development of new crop varieties include: Two new guar varieties, 'Hall' and 'Mills', developed by plant selection from P.I. 179930 and P.I. 263875, respectively, both from India. Both varieties possess high degree of disease resistance. 'Rancher' lupine is low in alkaloid content and is resistant to gray leafspot. It contains germ plasm of P.I. 189191 from Sweden. 'Magnolia' ryegrass, a new variety adapted to the southeastern United States, contains germ plasm from South America. Outstanding among its attributes is that of crown rust resistance. Plant introductions contributed to the new multiple-pest resistant variety of alfalfa, 'Washoe', developed by ARS and Nevada.

Industry Uses Acid-Modified Cereal Flours. Department scientists and engineers have developed a process for the chemical modification of cereal flours for use as surface-sizing agents for paper. The acid-modified flour, called AMF for short, was prepared in lots of several hundred pounds for a number of trials on laboratory, semicommercial, and commercial paper machines. Its performance on paper machines and the quality of the finished paper are about the same as with high-grade commercial modified starches. Encouraged by these findings, a wheat flour milling company with diminishing outlets for soft wheat flour, in cooperation with the Department and a commercial paper mill, conducted a trial in which laminated fiberboard was surface sized, the machine running at a normal rate of 75 tons per day. The finished fiberboard met the mill specifications. The cooperating milling company is planning to install a small plant to make AMF.

Four large milling companies are selling acid-modified corn or sorghum flour for use in paper sizing and gypsum board. All are using either the Department's process or a modification of it. Cost of the modified flours is favorable. No figures are available for current sales of AMF. The paper industry uses 600 million pounds of other corn and sorghum products per year for surface sizing and the gypsum board industry uses over 50 million pounds. Increased use of AMF is expected to develop largely in the coarse paper and box paper industry. The outlook is good, particularly in regions where flour and paper mills are in close proximity. Other producers of flour both in the U.S. and abroad are much interested in making AMF.

USDA's Wheat Gluten Process Commercialized. A continuous process was developed by Department scientists for separating wheat flour into starch and food-grade gluten. Five U.S. industrial installations are using this process, or modifications of it, on over 265 million pounds of flour annually. The process is simple in operation, and no addition of chemicals is required. A new plant now is nearing completion in the Pacific Northwest which will use second clears flour as the raw material. This plant will take advantage of regional markets and raw material supplies. The installation will include facilities to produce different grades of starch and food-grade gluten. Facilities for conversion of starch into sirup will also be installed. Gluten has many important uses including increasing the protein content of flours for baking and increasing the protein level of breakfast cereals. Wheat starch has broad use in foods and such industrial outlets as paper additives, wallpaper pastes, and plywood glue additives. Department scientists have been extensively consulted on the physical, chemical, and mechanical properties of wheat flour and its processing as a guide for design and installation of the new plant.

Food Protein Concentrates from Wheat Mill Fractions. Department research has shown that high-quality food ingredients can be made from bran and shorts, byproduct fractions produced when wheat is milled into flour. These byproducts have been sold heretofore as animal feeds, but now up to

50% of this material can be recovered as food-grade flours or upgraded feeds. In the new process, the bran and shorts are dried and remilled separately in conventional flour milling equipment. The resulting flours contain 25 to 40% protein and only 2 to 4% fiber. The biological value of the protein is high because it contains more than 4% of the scarce amino acid lysine. The flour milling industry has used these research results to develop a high-protein, general-purpose flour and a product that can be made into a nutritious gruel or beverage to supplement protein-deficient diets. Mill byproducts available are sufficient to allow recovery of nearly two million tons per year of these high-protein concentrates.

New Instruments for Rice Grading. A rice meter combining light reflectance and transmittance principles, has been developed to measure three of the important quality factors of milled rice; degree of parboiling, color or general appearance, and degree of milling. The instrument is also expected to be useful for continuous monitoring of degree of milling in rice plants.

Grain Thrower Causes Little Damage to Wheat. Studies to determine the extent and causes of breakage to grain by handling equipment are providing some significant, and often unexpected, results. For example when dry, cold wheat (11 percent moisture and 36° F.) was handled by the thrower under severe operating conditions, the maximum breakage in the wheat was only 0.3 percent. When such wheat was subjected to a handling treatment for as many as four times, the maximum breakage was about the same each time.

State Agricultural Experiment Stations

Fatty Acids of Wheat. Using gas chromatographic analysis, Minnesota biochemists have extended the knowledge of wheat lipids and comprehensively accounted for the distribution of fatty acids between free and bound lipids present in the milled fractions: bran, germ, and endosperm. Since bound lipids include the phospho- and glycolipids, one can predict that their fatty acid content is less than that for glycerides present in free lipids. In fact, the fatty acid content of free lipids is about twice that of bound lipids in bran and 1.5-fold greater in straight-grade flour. Linoleic acid is the predominant fatty acid in all fractions. Precision of the analytical method permitted an accounting for all the minor fatty acids in the range from 11-carbon to 22-carbon fatty acid molecules. Of the five major fatty acids in wheat lipids, their decreasing order of abundance is linoleic, palmitic, oleic, linolenic, and stearic; however, the distribution of the individual fatty acids between free and bound lipids is different for bran, shorts, and flour. Current interest in the intake of saturated fatty acids from a nutritional view is reflected in this work.

Improved Corn Proteins. To overcome the nutritional deficiencies which corn proteins possess, it has been a common animal feed practice to add enriching dietary supplements which compensate for the two limiting amino acids, lysine and tryptophane. Scientists at the Indiana Station have discovered in corn that mutant genes, such as opaque-2 and floury-2, change the protein composition and amino acid pattern of the kernel. A new high-lysine corn variety, opaque-2, containing 100 percent more lysine and 69 percent more tryptophane than normal seed, is being tested for high commercial yield and for nutritive value to swine, poultry, and humans. Another high-lysine corn, floury-2, contains both increased tryptophane and methionine levels. Improved protein quality in the new genotypes results from boosting the glutelin content, lysine-rich, and decreasing the zein level. Protein utilization of these genotypes for human nutrition offers distinct promise and has an important bearing on world food supplies.

Chemicals in Corn Control Resistance to the Corn Borer. Although larvae of the European corn borer have been responsible for losses to corn crops in past years amounting to over \$100 million per year, certain plant varieties have been found which are resistant to the corn borer. Recent basic research at the Iowa Agricultural Experiment Station has shown that a chemical substance present in the resistant strains of corn is a powerful feeding deterrent to the corn borer. The substance, isolated in pure form from corn seedlings and identified as a cyclic member of the hydroxamic acid family, is capable of killing the corn borer when fed at a level equivalent to its concentration in resistant inbred lines of corn. The possibility exists of field treating other varieties of corn with this substance to protect plants from corn borer ravages. Moreover, this finding may stimulate a search for chemical factors in natural resistance to other pests. A rapid method of analysis for specific hydroxamic acids in corn is being developed and will be of value to the plant breeder in predicting the corn borer resistance of new crosses.

"Milk" from Wheat Flour. Indiana scientists (partially supported by ARS-USDA) have prepared a milk-like product from wheat flour. Transferring a high level of protein from the flour to the milk-like product was the difficult problem. Using a process similar to digestion, the scientists were able to recover 85 to 90 percent of the wheat flour protein and to develop a dry high-protein concentrate which is soluble in water.

Nutritional properties of the product are similar to non-fat dry milk. When mixed with water, it looks like milk and has a bland flavor. The concentrate could also be added to soups and other prepared foods to improve their nutritional properties. The process was developed in search for a low-cost, palatable, milk-like product to improve human nutrition in distressed areas and underdeveloped countries.

I. FARM RESEARCH

BARLEY BREEDING, GENETICS, DISEASES, QUALITY, AND PHYSIOLOGY Crops Research Division, ARS

Problem. Barley is subject to yearly fluctuations in yield and quality due to factors such as climate, soil, diseases, lodging, winterkilling, shattering, drought, insect damage, and other factors. Barley diseases continue to cause great losses in barley. They lower the quality of the grain and restrict the area where high quality barleys can be grown. Additional basic work needs to be carried out to measure the degree of hybrid vigor that exists in this crop, on what effect hybrid vigor may have on malting and brewing quality, on the broad problems of disease and insect resistance, on additional genetic studies, and on field management practices to insure a reasonable cost of the seed. Commercial processing procedures for the production of malt and malt liquors are undergoing changes at the present time, and this raises the question of the suitability of presently used varieties for these purposes. The adequacy of the presently grown varieties needs to be determined, as well as to ascertain if other varieties may not prove superior with the new procedures. Increased efforts on basic research are needed in genetics, cytology, pathology, physiology, and breeding methods to provide information to strengthen the breeding programs, the work on quality improvement, and to provide a fuller understanding of the nature of winterhardiness, host-pathogen relationships, and the relation to molecular structure to gene action.

USDA AND COOPERATIVE PROGRAMS

The Department has a continuing long-range basic and applied program conducted by geneticists, pathologists, cytologists, physiologists, chemists, and agronomists, and related to the improvement of barley. Barley research investigations are conducted at Beltsville, Maryland, and in cooperation with the following State Agricultural Experiment Stations: Arizona, California, Idaho, Michigan, Minnesota, Montana, North Carolina, North Dakota, South Dakota, and Wisconsin. A Federal National Barley and Malt Laboratory is located at Madison, Wisconsin, and is operated in cooperation with the Wisconsin Agricultural Experiment Station. This Laboratory conducts quality research on barley for both State and Federal programs. The World Collection of Barleys is maintained and distributed from Beltsville. Federal personnel supervise four spring and four winter uniform nurseries.

Three P.L. 480 projects dealing with research on barley are in operation in three foreign countries: in Israel (terminated 1/20/66), on the screening and collection of cultivated barleys and related species, and breeding for disease resistance; in Poland, on finding sources of resistance to loose smut in barley and identifying genes for resistance; and in India, on the physiological studies of drought resistance in some crop plants.

The Federal research effort devoted to barley totals 17.4 professional man-years, of which 9.0 is for breeding and genetics, 3.2 for diseases, 4.0 for quality research, and 1.2 for physiology.

PROGRAM OF STATE EXPERIMENT STATIONS

A total of 35.1 professional man years is devoted to this area of research.

PROGRESS -- USDA AND COOPERATIVE PROGRAMS

A. Breeding and Genetics

1. Hybrid Barley. Further studies on the proposed scheme for hybrid barley announced last year, showed that about 2100 pounds of seed per acre can be anticipated from balanced tertiary trisomic plants when grown in the absence of competition with normal diploids. Seed from the balanced tertiary trisomic plants is that which will produce diploid male sterile plants for use in the crossing field to give the hybrid seed for commercial production. The amount of crossed seed produced using 8 different male parents and a common male sterile female ranged from 1700 to 2600 pounds per acre. These results are promising for the future of hybrid barley. Studies were initiated during the year on the feasibility of mechanically separating selfed seed set on balanced tertiary trisomic plants into two lots - one containing mostly trisomic plants and the other containing mostly diploids. Preliminary results indicate that such a separation is possible. This would greatly reduce the cost of using balanced tertiary trisomics in hybrid seed production. Several alternate schemes for producing hybrid barley seed also are being evaluated. Under California conditions, yield heterosis greater than 25 percent has now been observed in some crosses in which Blanco Mariout, Atlas 57, and Grande served as female parents.

2. Complementary Genes for Blue Aleurone. Conclusive evidence obtained this year shows that a blue aleurone color in barley is due to the complementary action of either two very closely linked transpositioned genes or to two sites in the same cistron also in transposition. On this hypothesis, two kinds of white barley should exist. Subsequently these have been found. This discovery, plus the possibility of electronically sorting blue seeds from white, and finding two male sterile genes closely linked with the above complex, hold the key to an alternate genetic system for hybrid barley. Under this scheme, plants heterozygous for the blue aleurone color and male sterile genes would be fertile and half of the seeds would be blue and half white. The seeds could be sorted for color, all white seeds would produce male sterile plants, and all blue seeds fertile plants. The latter would be heterozygous and used to maintain the stock to repeat the cycle.

3. Male Sterile Genes. More than 50 genetic male sterile genes have been collected by state and federal workers interested or working on hybrid barley. Twenty-two have been checked for allelism and 19 were found to be at separate loci. With such a high frequency of different loci, linkage with other genes which may be of value in the hybrid barley program should not be hard to find.

4. Unicm. Yield comparisons using a Unicm(nontillering) derived line showed the yields of Unicm were less than normal tillering lines at a low seeding rate, but that the reverse was true at higher seeding rates. The mean yields from all rates were 51.8 bu. for Moore, the tillering type, and 56.7 bu. for Unicm. Kernel weight and test weight were higher for Unicm.

5. New Varieties Released. Two improved varieties were released to seed growers. The spring, two-rowed variety Hypna was released jointly by the Montana Agricultural Experiment Station and the Crops Research Division, ARS, U. S. Department of Agriculture. Hypna was developed jointly by these two agencies from a cross of Glacier x Compana. Hypna is 20% taller, 4% higher yielding, more lodging resistant and has larger seed than the variety Compana, which it is intended to replace. It should be adapted to dry land production in the Northern Great Plains area.

Godiva was developed and released jointly by the Utah State University Agricultural Experiment Station and the Crops Research Division, ARS, U. S. Department of Agriculture. Godiva is a spring, hull-less variety and is especially adapted to fertile irrigated lands of the intermountain region. Godiva does not perform well on dry land or on soil of average fertility. After allowance is made for lack of hulls, Godiva outyields the commercial varieties Trebi and Bonneville and is more resistant to lodging. Godiva also has resistance to some diseases common in the area, and threshes easily but must not be threshed too vigorously, or germination may be lowered.

6. Cereal Leaf Beetle. The level of resistance in the World Collection of Barleys to the cereal leaf beetle (Oulema melanopa) is not as good as that for wheat; but is similar to that of oats. Crosses have been made between the most resistant entries in the World Collection and adapted cultivated varieties, and between resistant entries, in order to produce lines which will combine the genes for resistance. Progenies from these crosses will be studied to determine the inheritance of resistance. The wild species of barley tested thus far offer a higher level of resistance than the cultivated types.

B. Diseases

1. Virus Diseases. The application of methods for increasing and maintaining seed free from the barley stripe mosaic virus has reduced the losses from this virus, which was the most important disease on barley in the United States from 1951 to 1960. In 1965 less than 2% of the commercial barley fields surveyed in North Dakota had plants with the virus.

The yield of the variety Moreval was reduced by the inoculation with virus strains to which the variety was immune. The virus in the seed was not inactivated by heating dried seed to 130°C for 30 minutes, alternate freezing and thawing, or soaking and drying. Plants resistant to the barley stripe mosaic virus were found to have less virus than susceptible plants. A rapid, efficient virus purification method has been developed by using chloroform and charcoal.

Temperature affects the development of the barley yellow dwarf virus disease. The virus progresses through the plant more rapidly at 60° and 70° F than at 50° or 80° F. The increase in respiration of the infected plants was greater at 60° and 70° F than 50° or 80° F. The growth of the second, third, and fourth leaves of seedlings was shown to be correlated with the age of seedling at time of inoculation. The younger the leaf when inoculated with the virus, the greater the reduction in growth.

2. Barley Rusts. A new physiologic race of the barley leaf rust fungus, virulent on many commercial winter barley varieties, was identified. Varieties resistant to the new race have been found and are now being incorporated into the breeding programs.

The complete life cycles of the two rusts on barley caused by Uromyces and Puccinia were observed in nature in Israel (PL 480 research). Cultures of Puccinia hordei from Hordeum bulbosum do not attack Hordeum vulgare, and cultures of Puccinia hordei from Hordeum vulgare do not attack Hordeum bulbosum. Five species of Ornithogalum are known to be alternate hosts of Puccinia hordei. Some of these species are specific as to the species of barley to which the fungus will attack.

3. Powdery Mildew. A method of inoculating a plant with 3 or more cultures of powdery mildew has been developed. Small cross-sections of leaves are floated on benzimidazole solution in 2 x 2 cm plastic chambers, separated into groups as desired, inoculated, and then incubated with the individual chambers covered. Up to 5 sections from one leaf can be tested.

In research under PL 480 in Israel, cultures of the powdery mildew fungus were isolated which attack only one genera, while others were isolated which attack more than one genera. The latter cultures may have resulted from crosses between cultures which attack different genera.

4. Net Blotch. Barley varieties resistant to the net blotch fungus have been identified. A method has been developed for crossing the fungus, which can be used for studying the sexuality and other characteristics of that fungus.

In PL 480 research in Israel, cultures of the net blotch fungus found in that country were heterothallic and sexually compatible with cultures from North America. Monoconidial isolates of the net blotch fungus were pathogenically unstable and retained their variability after five passages through barley. Varieties of cultivated barley and barley species resistant to the net blotch fungus have been collected from fields in Israel.

C. Quality

1. Effect of Cultural and Environmental Practices on Quality. In 1964, the high malting quality upper midwestern varieties, Traill, Trophy, and Larker, were grown successfully in Michigan's upper peninsula and at Tule Lake, California. Traill, however, maintained a higher malting quality than the other two under these widely different environmental conditions.

In continuing studies at Wisconsin, spraying growing barley with fungicides improved barley color and had no other influence on quality. The best barley quality resulted from the earliest planting date and second or third harvest date.

2. Effect of Gibberellic Acid on Malting. Malting cage rotation speeds higher than 960 rph are required to reduced rootlet development. Fast rotation combined with gibberellic acid caused excessive acrospire growth and low malt recovery.

3. Varieties from State and Federal Breeding Programs. Almost 3,000 barleys from the 1964 crop were analyzed and about 2,800 were malted. More than $\frac{2}{3}$ were from the western states. Fifty-six advanced selections were compared with checks by pilot brewing and 70 were evaluated by micro-brewing. In evaluation of commercial Dickson samples, malting and brewing performance was similar to Trophy and Larker. Promising 6-row selections from breeding programs at North Dakota, Minnesota, Michigan, and Wisconsin appeared to be nearing the final testing before release. Most promising 2-row selections were from Pullman, Washington, and Aberdeen, Idaho.

4. Emetic Compound. Emetics from scabbed grain and culture filtrates showed some separation in thin layer, column, and gas chromatography. The latter technique especially justifies further study.

5. Enzymes. The release of alpha-amylase from aleurone tissue to incubation medium in the presence of gibberellic acid is much slower than production of enzyme in the tissue. The malt peptidase (Bapa-ase) while present in aleurone tissue, does not increase during incubation.

The proteases of germinated barleys were separated into five fractions by column chromatography and gel filtration. Preliminary study of the fractions indicated the presence of three peptidases and three proteinases that appear to be distinct enzymes.

6. Amino Acids. Small differences in amounts of free amino acids in ten barley varieties were found, but the variety Olli contained larger amounts of most acids and especially proline. The better malting varieties tended to be higher in free amino acids than those of poorer quality.

D. Physiology

1. Cell-Wall Polymers Interfere with Ice Lattice to Prevent Freezing Damage. Polymers isolated from the cell walls of hardy and nonhardy cereal plants were found to consist mainly of polysaccharides. These polysaccharides contained mostly xylose and arabinose as monomer units. The effects of these polymers on equilibrium and kinetic relationships which occur during freezing of water films over cellulose were evaluated, and a thermodynamic technique was derived for studying freezing inhibitors extracted from cell walls of hardened winter cereals. The investigations also included commercially available polysaccharides and frozen food stabilizers, xylans extracted from a winter-hardy pond weed, and xylans from winter cereals grown under a wide range of environmental conditions. A unique fractionating procedure was developed for classifying polysaccharides based on the interaction of the polysaccharides with an expanding ice lattice. It is hoped that the identification of specific molecular features with specific types of freezing inhibition will, in the end, be amenable to genetic control.

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CORN AND SORGHUM BREEDING, GENETICS, DISEASES, PHYSIOLOGY, AND CULTURE
Crops Research Division, ARS

Problem. During the last 25 years, the hybrid seed industry has provided an increasing percentage of the hybrid corn and sorghum seed planted by farmers. This development has permitted a reorientation of our program to give increasing emphasis to basic research in genetics, physiology, and pathology. Need for increased activity in these areas has long been recognized but was not possible as long as primary emphasis was required in developmental breeding. The present division of responsibility and effort should permit increasing efficiency. Research must be a continuing activity; new problems requiring solutions are constantly arising and many old problems have never been effectively resolved. Typical of the new problems is the recent serious outbreak of corn viruses; maize dwarf mosaic in the Corn Belt and corn stunt in the southern states. Recent studies indicate that corns may be developed having a much more favorable amino acid balance. Can such genetic traits be effectively incorporated into commercial corn thereby increasing the efficiency of livestock production and enhancing the value of the crop in production of human food? Older problems, still unresolved, include a completely satisfactory explanation of heterosis in either genetic or physiologic terms. Additional information is needed on genetic control of biosyntheses of oil, protein, carbohydrate, and other important constituents to permit the development of types having increased industrial or nutritional potential. The chemical basis for resistance to disease and insect pests, and the host-parasite interaction system remain largely unexplored.

USDA AND COOPERATIVE PROGRAM

The Department has a continuing long-term program involving geneticists, physiologists, pathologists, and agronomists engaged in basic and applied studies relating to the improvement of corn and sorghum. Corn research is conducted at Beltsville, Maryland and Charleston, South Carolina; and in cooperation with State Agricultural Experiment Stations at Tifton, Georgia; Urbana, Illinois; Lafayette, Indiana; Ames, Iowa; State College, Mississippi; Columbia, Missouri; Raleigh, North Carolina; Wooster, Ohio; Brookings, South Dakota; Knoxville, Tennessee; and Madison, Wisconsin. Sorghum research is cooperative with State Agricultural Experiment Stations at Manhattan and Hays, Kansas; Lincoln, Nebraska; Stillwater, Oklahoma; and Chillicothe and College Station, Texas. Research on improvement of corn, sorghum, and millet also is being conducted in Africa for the Agency for International Development.

Seventeen P.L. 480 projects dealing with corn and sorghum research are in operation. In India, projects deal with corn and sugarcane genetics; basic research on carbohydrate metabolism; the genetic effects of radiation; techniques to increase the storage life of pollen; storage, maintenance,

and distribution of millets germ plasm; cataloguing and classifying genetic stocks of sorghums; responses of various germ plasm sources to agronomic practices; investigations on diseases of sorghum and other important millets; research on maize diseases with special reference to Erwinia carotovora var. zeae; chemical composition of pearl millet; and studies on Helminthosporium species on corn and sorghum. In the Philippines, there is research on downy mildew of corn; in Spain, on factors affecting the frequency of monoploid maize; in Yugoslavia, on collection, classification, evaluation and preservation of local germ plasm; and on leaf redness in corn; and breeding corn for high oil and protein content; and in Brazil, on evaluation of Latin American corn germ plasm for U. S. use. The following cooperative agreements are in effect: Missouri, with the Entomology Research Division, resistance to the corn earworm; Nebraska, quantitative genetic studies with sorghum; and Illinois, genetic control of fatty acid synthesis. Contract research is being done in Arizona on resistance to the charcoal rot disease of sorghum.

The federal research effort in the United States devoted to corn and sorghum totals 36.1 professional man-years. Of this, 20.4 are devoted to breeding and genetics; 7.5 to diseases, and 8.2 to culture and physiology. In addition, 5 professional scientists conduct research in Africa on corn, sorghum and millet under funds provided by AID. Of these, 3.7 are devoted to breeding and genetics, and 1.3 to diseases.

PROGRAM OF STATE EXPERIMENT STATIONS

A total of 145.8 professional man years is devoted to this area of research.

PROGRESS -- USDA AND COOPERATIVE PROGRAMS

A. Breeding and Genetics

1. Corn Genetics. Basic genetic studies are underway at Ames, Iowa; Beltsville, Maryland; Columbia, Missouri; and Raleigh, North Carolina. Research involves three separate areas: classical genetics and cytogenetics, quantitative genetics, and biochemical genetics. These have the common objective of a more complete understanding of the principles underlying improvement of corn.

a. Classical Genetics and Cytogenetics. Several mutation systems are known in maize. While these have quite different characteristics the behavior of each can be accounted for by the assumption of an episome-like particle. Such a particle is not a normal chromosomal constituent but except for occasional transposition or loss appears to be under chromosomal control. The conversion phenomenon at the B locus as well as "ratio distortion" and other mutagenic effects of a plant virus appear to be of this nature. Chromosomal aberrations may have a pronounced effect on crossing-over. Detailed studies, involving "abnormal 10" are designed to measure the influence of crossing-over on estimates of genetic variance.

High haploidy appears to be under genetic control but the details of inheritance are not yet clear. Studies supported under a PL 480 grant in Spain have given no clear evidence for the modification of haploid frequency through treatment of either egg or sperm.

b. Quantitative Genetics. Various models have been utilized to estimate the relative importance of different types of gene action involved in heterosis. Additive and dominance effects are normally found; the relative importance of each varies with the population and trait under consideration. No evidence has been found indicating the importance of epistatic effects in random-mating populations. Epistatic effects have been detected, however, in comparisons involving single-cross and three-way cross hybrids. If epistasis is of importance at the hybrid level none of the breeding systems in current use would have high efficiency. New methods are being developed which would permit greater emphasis on epistatic gene effects. Several types of recurrent selection systems are being evaluated in the United States plus in a PL 480 project in Brazil. Within the limited genetic base available in the United States, rates of gain have been achieved. Quantitative genetic theory has been based largely on intra-population considerations. Theory is currently being expanded to cover inter-varietal populations.

c. Biochemical Genetics. Bacteria, fungi, yeasts and some algae have been extensively used in biochemical genetics. Relatively little information is available from higher plants although mutant types having specific biochemical properties are important items of commerce. Biochemical genetic studies are currently underway on carbohydrate, oil, carotenoid and anthocyanin synthesis. Extensive genetic data are available on the inheritance of differences among carbohydrates, carotenoids and anthocyanins but only limited information is available on biosynthesis. Work is furthest advanced on anthocyanin pigmentation of the aleurone. Here the sequence of action of several genes has been established through paired comparisons of developing aleurone tissue but the specific compounds accumulating at each such block remains to be established. A beginning has been made on the biosynthesis of carotenoid pigments. Genetic stocks are available which modify the type of carbohydrate stored in the endosperm. Enzyme and radioactive isotope studies are underway to determine the biosynthetic pathways. Tissue culture studies are being explored. If adequate techniques can be developed this approach will provide a continuing supply of tissue at a developmental stage when carbohydrate deposition is progressing most rapidly.

2. Dent Corn Breeding.

a. Inbred Lines Released. Eighteen inbred lines were released from cooperative breeding programs. The lines released from Iowa were B59, B64, and B65 and pollen restorer versions of B42 and B45. B59 and B65

have moderate to good resistance to the first brood of the European corn borer and B6⁴ has a high level of tolerance to the western corn rootworm. The five lines, GT112D, L578D, Mp462D, Mp44D and T294D, were released from State College, Mississippi. These are modified dwarfs and, in hybrid combinations, give short-statured hybrids equal in yield to their normal-statured counterparts. Eight lines, four white and four yellow were released from Knoxville, Tennessee. These were T218, T220, T222, T224, T315, T331, T357, and T393. Inbred lines T222 and T224 possess resistance to the corn virus occurring in Tennessee.

3. Sorghum Genetics. Genetic and breeding studies are underway at each of the locations where cooperative work is in progress. Cytogenetic studies are concentrated at College Station, Texas; disease resistance at Manhattan, Kansas and College Station, Texas; and the genetics of morphological and physiological traits at Hays and Manhattan, Kansas, and Lincoln, Nebraska. Ten primary trisomes of sorghum have now been isolated. Some progress has been made in the association of a trisome with a particular chromosome of a translocation heterozygote. When these studies are completed it will be possible to assign linkage groups to known chromosomes. Forty-two homozygous translocations now available are known to involve at least seven of the ten chromosomes. Resistance to Periconia (milo disease) appears to be a mutable locus. The mutation rate is approximately doubled in the presence of the organism and the toxin which it produces. Test crosses to establish the identity of these mutations have been made and experiments designed to measure the frequency of back-mutation. One of the loci affecting plant height is also unstable. A series of tall and dwarf segregates have been obtained from presumed dw₃ → DW₃ mutants in preparation for detailed tests of allelism and heterotic response.

4. Sorghum Breeding. Two sorghum lines were released. The line 4610 was released at Lincoln, Nebraska. It is a pollinator line derived from the cross Western Blackhull x Day. The second line, KS7 was released at Manhattan, Kansas. Approximately half of the World Sorghum collection was grown at Isabela, Puerto Rico, for seed increase and storage at the National Seed Storage Laboratory at Ft. Collins. An additional generation of backcrossing was obtained in the conversion program which is designed to introduce desirable height and maturity genes into the most promising items from the World Sorghum collection. Seed for the increase came from a PL 480 project in India.

B. Diseases. The more important diseases of 1965 were the corn viruses and a severe epiphytotic of Gibberella in the northern part of the Corn Belt.

1. Corn Diseases

a. Viruses. At least three virus diseases have been identified; corn stunt, maize dwarf mosaic, and a wheat streak mosaic-like type which causes red kernel striping. Corn stunt has been confirmed by transmission tests in Louisiana, Mississippi and Florida. The known vectors are species of the leaf-hopper, Dalbulus. Other vectors must be involved, however, as the disease becomes severe before the known vectors are present. Maize dwarf mosaic was more widely distributed than in previous years but with slightly reduced losses. This disease is most severe, but not exclusively limited to areas where Johnson grass is a common weed. Resistance has been found to both of these viruses but additional sources are necessary. A third virus has been found with heaviest concentration in Ohio, Indiana, Michigan, and Wisconsin. This virus resembles wheat streak mosaic and can be transmitted mechanically or by a mite. In addition to the ephemeral mosaic symptoms a red striping develops on the kernels. Estimates of yield losses are not available but appear to be much less than for maize dwarf mosaic. The relation of these viruses to a disease known as "leaf redness" in Yugoslavia remains unknown. The causal agent of the "leaf redness" is not yet known. (PL 480 research)

b. Gibberella Ear Rot. Red kernel striping and Gibberella ear rot occurred in the same geographical area. Feeding trials have indicated that the red striping disorder has no deleterious effects on acceptability or rates of gain. The Gibberella infected material is avoided by swine. When forced to eat such infected grain, gains are drastically limited.

c. Corn Rusts. Sources of resistance are now available to the known races of Puccinia sorghi and P. polysora. Several loci are involved. Linkage relations of several of the P. sorghi resistance genes has been established. Less information is available for resistance to P. polysora. Preliminary data suggest that all loci conditioning resistance are carried on the same chromosome.

d. Downy Mildew. This disease appears to be increasing in importance in South Texas. Resistance has been found to strains occurring in the Philippines (PL 480 project) and such sources of resistance are under observation in Texas.

e. Helminthosporium. Work has been directed toward an understanding of population structure, the inheritance of pathogenicity and host-parasite relations. Crosses between two isolates of Helminthosporium, one pathogenic to Calamagrostis and the other to Phalaris, indicate that pathogenicity to each host is controlled by a different gene. When the same isolates are tested on 7 additional graminous species resistance to four hosts is inherited independently and the remaining three are linked. In other crosses with a conidial morphology similar to H. maydis 91

isolates were tested against 30 gramineous hosts. Isolates ranged in pathogenicity from ability to attack 4 species to those attacking 25. Genes conditioning victoxinine production are found in both pathogenic and non-pathogenic isolates of H. victoriae while only pathogenic isolates produce the host-specific toxin. Studies are underway to determine whether the genetic blocks in the sexual cycle to perithecial, ascus and ascospore formation can be repaired by growth extracts and steroid compounds. Quantitative estimates were made of the rhizosphere mycofloras of three inbreds; two susceptible to stalk and root rot and one resistant. The susceptible inbreds supported the largest populations although there was considerable seasonal fluctuation corresponding to the growth phase of the host.

2. Sorghum Diseases. Downy mildews (Sclerophthora macrospora and Sclerospora sorghi), head smut and charcoal rot were the most serious diseases of sorghum in 1965.

a. Downy Mildews. Sclerophthora was widespread in Kansas in 1965. Yield losses up to thirty percent have been reported in low-lying fields. The above normal incidence of this disease appears to have been related to above average rainfall. Techniques have now been developed for the ready identification of the disease. Sclerospora sorghi has increased in distribution each year since its discovery in 1961. Thus far this disease is primarily confined to Texas but is slowly progressing northward.

b. Head Smut. Losses in yield from head smut ranged from highs of 20 to 30 percent in south Texas to one to five percent in Kansas. Losses could be materially reduced by a fuller utilization of resistant types. Breeding for resistance has been hampered somewhat by inadequate techniques for artificial infection. Germination of chlamydospores is often irregular resulting in low and erratic levels of infection. Thus several years of testing may be required before relative ratings can be assigned to any but the most susceptible entries. Extensive studies on maturation of chlamydospores in relation to germinability are under way.

c. Charcoal Rot. A contract with the University of Arizona supports the development and field evaluation of techniques to measure relative resistance to this disease. It has been established that the conditions required for the development of the disease include (1) plants in milk to dough stage; (2) daily maximum temperature above 35°C; and (3) a sub-lethal moisture stress. The most difficult element to control is moisture stress. In general early types are more susceptible than later material. Extreme susceptibility can be readily identified but no highly resistant types have yet been observed.

C. Physiology and Culture

1. Corn.

a. Protein Synthesis. Attempts have been made to study the incorporation of radioactive amino acids into protein through use of protein bodies, microsomes and ribosomes. The apparent incorporation was low and has been interpreted as an absorption phenomenon. Bacterial contamination has been a real problem. Many previous studies on incorporation are suspect through failure to measure or control bacterial incorporation.

b. Fat Synthesis. Collections were made 10, 15, 20, 30, and 45 days after pollination for 5 strains of corn. The total lipid extracts were fractionated into sterols esters, triglycerides, free fatty acids, sterols, and phospholipids by thin-layer and column chromatography. Eighteen individual components were found. Phospholipids made up 50% of the lipid extract in the 10-day sample but only four to seven percent at maturity. The fatty acid composition of the lipids changed markedly as the grain matured.

c. Carbohydrate Synthesis. A considerable number of mutant stocks are available which influence the amount and characteristics of the reserve carbohydrates in the corn endosperm. One or more of the enzymes involved appear to be bound to the starch granule (PL 480 research). It is postulated that the conversion of sucrose to starch involves at least four enzymes: sucrose synthetase, UDPG pyrophosphorylase, ADPG pyrophosphorylase, and starch synthetase. The biosynthetic pathways may be somewhat "leaky" as sh₂ and bt₂ lack ADPG pyrophosphorylase but produce 25-30% as much starch as normal corn. Radioactive labeling indicates that mature and newly synthesized or immature starch may be separated on the basis of differential solubility in DMSO (dimethylsulfoxide) and CaCl₂. DMSO extracts the bulk of the mature starch and CaCl₂ the newly synthesized starch.

d. Mineral Nutrition. IaB8 and IaB8A are near isogenic lines which differ in response under conditions of magnesium deficiency. When grown in solution culture at varying levels of magnesium it was found that IaB8 contained a higher magnesium content in roots, nodes, and internodes but a lower magnesium content in the leaves. Thus IaB8A, the strain exhibiting magnesium deficiency symptoms, had the higher magnesium concentration in its leaves. Low levels of mineral nutrition are associated with low levels of organic acid. A reverse relation was noted between the accumulation of magnesium and phosphorus. No explanation is yet available for the development of deficiency symptoms.

2. Sorghum. Physiologic studies with this crop are concerned with water stress and its direct and indirect influence on various plant processes.

A microwave refractometer has been developed, under a cooperative agreement, and is currently being evaluated to establish the feasibility of measuring water content of intact plant material. In the meantime studies on soluble carbohydrates were undertaken to provide essential background on translocation patterns of assimilates at different growth stages and on regrowth rates in forage-type sorghums.

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WHEAT AND RYE BREEDING, GENETICS, DISEASES, QUALITY,
PHYSIOLOGY, AND CULTURE
Crops Research Division, ARS

Problem. Wheat is the most extensively consumed human food crop grown in the United States and ranks equal with rice on a world-wide basis. Wheat is grown commercially in 42 states, and to some extent in five others. Rye is grown in 31 states with sparing use made of it in some others. This obvious diversity in cultural environment is accompanied by a very diverse range of problems of adaptation, disease and insect pests, and of utility of the crop, including milling and baking quality. The problem, therefore, is to develop suitable practices, improved varieties, and better pest controls, to maintain or improve the quality of the crop produced, and to develop new knowledge as a basis for further improvements. This requires a broad range of disciplines in the scientists who conduct the work, and it requires a number of locations in order to be situated where the specific work can be done most effectively.

USDA AND COOPERATIVE PROGRAM

The Department has a long-term program of research leadership in wheat investigations. The objectives of the research are to stabilize production by reducing losses, to increase efficiency of production, to improve the quality of wheat wherever grown in the United States, and to accumulate and disseminate knowledge. Basic and applied research is performed in the laboratory, greenhouse, or field, and involves the sciences of agronomy, pathology, chemistry, entomology, physics, and statistics. Regional and national leadership is given to several phases of the work.

Federal research is concentrated at eight centers, but other locations are required either to provide proximity to the problem or to take advantage of facilities and contacts with cooperating workers at such locations. At Beltsville, Maryland, research is conducted in breeding and genetics, diseases, and physiology. In cooperation with state agricultural experiment stations, research in breeding and genetics is conducted at Mesa, Arizona; Tifton, Georgia; Aberdeen, Idaho; Lafayette, Indiana; Manhattan, Kansas; St. Paul, Minnesota; Columbia, Missouri; Bozeman, Montana; Lincoln, Nebraska; Fargo, North Dakota; Stillwater, Oklahoma; Corvallis, Oregon; Pendleton, Oregon; Brookings, South Dakota; College Station, Texas; and Pullman, Washington. Research in disease work is conducted at Tifton, Georgia; Aberdeen, Idaho; Urbana, Illinois; Manhattan, Kansas; St. Paul, Minnesota; Bozeman, Montana; Lincoln, Nebraska; Fargo, North Dakota; Stillwater, Oklahoma; Corvallis, Oregon; Mayaguez, Puerto Rico; Brookings, South Dakota; College Station, Texas; and Pullman, Washington. Research on quality is conducted at Manhattan, Kansas; Fargo, North Dakota; Wooster, Ohio; and Pullman, Washington. Research in physiology and culture is conducted at Bozeman, Montana; Lincoln, Nebraska; Corvallis, Oregon; Brookings, South Dakota; and Pullman, Washington.

There are 51.0 professional man-years assigned to this area, of which 17.0 are for breeding and genetics, 18.7 for diseases, 13.0 for quality research, and 2.3 for culture and physiology. During the year 10 grants and special cooperative agreements were in force or initiated. These concerned basic research on diseases in North Dakota, Nebraska, and Pennsylvania; basic genetics and cytology in Nebraska and Idaho; basic plant physiology of insect resistance in Oklahoma; nitrogen reductase in Illinois; fungus inhibitors in Oregon; and fundamental studies of quality factors in North Dakota and Montana.

Ten P.L. 480 projects on wheat and rye include the following: in Spain, polyploidy in rye, and cereal rusts; in Egypt, cereal rusts and smuts; in Pakistan, cereal rusts and smuts; in Poland, pathogenic diversity in rust; in Israel, root systems, and origin of cereals; in India 3 rusts of wheat; in Yugoslavia, leaf rust resistance, and branching spike types of wheat.

PROGRAM OF STATE EXPERIMENT STATIONS

A total of 81.5 professional man years is devoted to this area of research.

PROGRESS -- USDA AND COOPERATIVE PROGRAMS

A. Breeding and Genetics

1. New Varieties and Germ Plasm Released. 'Leeds', a new durum variety, is superior to Wells and Lakota, currently most popular, in test weight, kernel size, and stem rust resistance. It resembles present leading varieties in yielding ability, height and maturity. It was developed in cooperation with the North Dakota Agricultural Experiment Station. The larger seed size is especially important both for domestic and foreign markets.

'Sheridan' is a hard red spring variety developed in cooperation with the Montana Agricultural Experiment Station. The variety is resistant to common races of leaf, stem, and stripe rust; it also resists stinking smut and scab. Higher yield and quality are other advantages over varieties it will replace.

'Fortuna' is the first solid-stemmed hard red spring variety to combine wheat stem sawfly resistance with adequate leaf and stem rust resistance, early maturity, good yield, and good quality. It was developed by the North Dakota Agricultural Experiment Station and the Crops and Entomology Research Divisions of ARS. The variety is limited in usefulness to the sawfly infested areas.

'Sawmont' is a wheat stem sawfly resistant winter wheat developed by the Montana Agricultural Experiment Station in cooperation with the Crops and Entomology Research Divisions of ARS. Sawmont has more winter hardiness than Rego, the only other available resistant variety of winter wheat.

Agronomic and quality properties appear satisfactory.

'Hume' was developed in South Dakota and tested and released in cooperation with ARS. It is an early maturing, hardy, stem rust resistant variety of hard red winter wheat.

'Sturdy' is a hard red winter variety developed in Texas in cooperation with ARS. It is the first semidwarf variety of this market class. Somewhat lacking in hardiness, it is expected to be limited mainly to Texas.

'McCall and 'Wanser' are hard red winter wheats developed jointly by the Washington Agricultural Experiment Station and ARS. McCall has more snow mold tolerance and better emergence properties than other varieties currently available. Wanser has resistance to stripe rust in the adult stages. Both have good bread-making quality when the protein level is in the suitable range.

'Lemhi 66' is a stripe rust resistant form of Lemhi 53, a soft white spring wheat. It was developed in cooperative work by the Idaho Agricultural Experiment Station and ARS. It resembles Lemhi 53 in all important ways and was developed in a crash program to provide farmers with a rust resistant variety.

'Nugaines', a soft white winter wheat, was released as an improved milling type of Gaines. It is a semidwarf variety with about the same properties as Gaines except for higher test weight and better milling. The variety was developed by ARS and the Washington Agricultural Experiment Station.

'Moro' is a club wheat developed to replace Omar. The new wheat has demonstrated high resistance to stripe rust at all stages of growth and resistance to all known races of both common and dwarf bunt. It has weaker straw than Omar and may shatter more. It was developed by the Oregon Agricultural Experiment Station in cooperation with the Agricultural Research Service.

'Riley', soft red winter wheat, was developed cooperatively by ARS and Purdue University. Riley is resistant to loose smut and leaf rust, and has a combination of other resistances and good qualities. It has shown exceptionally high yield.

'Benhur' is a variety of soft red winter wheat released to combat the new biotype of hessian fly, race B, that attacks Monon, Riley, and certain other eastern wheats which derive their resistance from W38. The variety is the result of joint work by the Entomology and Crops Research Divisions of ARS and a team of Purdue University Scientists. The variety carries resistance to leaf rust, also.

2. Cereal Leaf Beetle Resistance. During two years of testing in Michigan, about 42,000 lines of wheat, oats, and barley have been rated for resistance. In the current year 7,150 wheats were rated; many were retests of promising lines from the previous years' tests. Of 179 winter wheats retested, 38% had low damage; of 266 durum spring wheats retested, 6% were resistant; and of the common spring wheats retested, 14% were resistant. Triticum-Agropyron derivatives yielded one resistant entry among 831 tested. Approximately 500 wheat varieties selected for cereal leaf beetle resistance under field nursery conditions were tested for resistance in the laboratory. Utilizing tests of growth response and survival of first instar larvae and ovipositional preferences, 18 spring wheat varieties had high levels of resistance to both the larval and adult forms of the beetle. Plant pubescence of a specific type was coincidental with the highly resistant wheat varieties. Winter wheat varieties which were immune to beetle attack for two years in fall-planted field nurseries were susceptible when tested as non-vernalized seedlings in the laboratory. Cooperative with the Entomology Research Division and Michigan State University. Resistance has been introduced into genetic and breeding studies in Michigan, Indiana, South Dakota, and elsewhere.

Explorations and seed exchange programs with the Balkan countries were stepped up during the year to obtain old land varieties that may possess beetle resistance. (Cooperative with New Crops Research Branch). Species related to wheat were propagated in Idaho to facilitate testing under beetle outbreaks (Cooperative with Idaho Agricultural Experiment Station).

3. Hybrid Wheat. Male sterile and fertility restoration lines are being developed in spring, winter, and durum wheat varieties. Domestic varieties and introductions are being evaluated as sources of fertility restoration. Of five spring wheats tested, Rescue and Thatcher produced the largest amounts of pollen. In a 10-parent diallel study of combining ability of durum wheat lines F_1 hybrids ranged from about 92% to 184% of the higher parent of a particular cross. However, the highest yielding F_1 outyielded Wells, the predominating durum variety, by about 15%. Further research will have to be made for higher yielding combinations if hybrid durum is to succeed. Several sterile and mostly sterile lines derived from original crosses of durum onto Triticum timopheevi have been crossed four times with Marrocos 9623. Some lines were completely sterile at each backcross, and others were partially fertile under certain conditions. A male sterile line derived from Bison male sterile (Nebraska source) has been crossed four times with Marrocos 9623. These lines have shown complete sterility.

In crossing blocks of hard red winter wheat the degree of ergot infection increased as the disparity between blooming dates of the parent lines widened. In the most extreme cases, 60 percent of the harvested product was in the form of ergot bodies.

The status of hybrid wheat is that primary increases of male sterile stock are being made to permit field-scale testing of seed set in male sterile and crossing fields. The latter will provide seed for critical field trials of heterosis from seed produced by the male sterile system.

4. Wheat and Rye Genomes Related. It was established that rye chromosome III belongs to homoeologous group 2 in wheat. It has been successfully substituted for chromosome 2A and 2B in Missouri.

5. Tetraploid Rye Synthetics Show Promise. Based on the suitability of the tetraploid rye for competing in yield and quality with the diploid one and on some good results already obtained by mixing inbred lines, it is expected that by means of polyploidy and inbreeding, synthetic varieties of tetraploid rye might be obtained that will exceed the present commercial varieties. Statistical analyses show that the polycrossed tetraploid material yields more than the original diploid one. These results confirm the hypothesis on which this rye breeding project was based: Polyploidy and inbreeding simultaneously applied in a rye breeding program are effective. (P.L. 480 research in Spain)

6. Chromosome 6B Map. An experiment conducted in 1965 provided the first reliable data for Co and at the same time produced slight modifications in some of the other distances. The map is now as follows: Lr9 0.09 Sr11 10.0 Ki 48.5 B2 0.43 centromere 53.7 Co.

It must be emphasized that the values between Ki and B2 and between B2 and Co are small, because there are doubtless double crossovers in these long intervals that reduce the apparent map distance. Between Lr9 and Sr11 the very short map distance presumably does not reflect a similarly short physical distance, for the Lr9 umbellulata segment unquestionably reduces crossing over greatly in the region adjacent to it.

7. Stem Solidness May Be Extended. Approximately 240 plant rows of F_4 interspecific hybrids with solid stems were observed in Montana. Selection was made of 110 lines, consisting of 57 of Yogo x Triticum pyramidale, 20 of Yogo x T. turgidum, 13 of Yogo x (T. polonicum x T. turgidum), 12 of Yogo x T. orientale, and 8 of $BC_3 F_4$ Yogo x T. polonicum.

Parental and F_2 populations of 9 foreign wheats crossed to Rescue were evaluated for stem solidness. Rescue was most solid in 5 crosses, but least solid in crosses with 4 foreign introductions, (P.I. numbers 191845, 94585-5, 94585-12, and 191835). Wheats that are more solid than Rescue may be of some value in the sawfly breeding program in Montana and North Dakota.

8. Introgression Between Aegilops Polyploids Seen as Bridge to Wheat Species. Spontaneous and artificial hybrids were studied for genetic transfer of characteristics and chromosomal affinity (P. L. 480 research in Israel). Species of aegilops sharing one genome are loosely and interconnected as revealed by occasional pairing in first generation hybrids and improvement in pairing in later generations with an increase also in fertility.

9. Monogenic Durum, and Common Wheats Derived for Stem and Leaf Rust, and Mildew Resistance. A series of backcrosses have been completed at Beltsville transferring single genes for mildew resistance to hexaploid Chancellor wheat from Khapli and Yuma tetraploids. The resistance of durum varieties Langdon and Ld 375 to culture 15-WL (race 15) of stem rust is assumed to be derived from Khapli emmer. Tests of F_3 seedlings from Langdon x Mindum and Langdon x Wells indicated that the high degree of resistance of Langdon to culture 15-WL is conditioned by a single gene. Eleven homozygous resistant lines derived from C.I. 8155, seven from P.I. 94701, and six from P.I. 192168 were isolated from F_4 and backcross- F_3 families. The lines appeared to have single genes for resistance in F_3 , F_4 , backcross- F_2 , and backcross- F_3 tests.

At Fargo, North Dakota, three lines, each monogenic for resistance derived earlier from the hexaploid wheat Reliance, were crossed in combinations with three monogenic lines derived from Marquis. Each of the monogenic lines from Reliance and from Marquis was crossed to Little Club and backcrossed to Little Club. Two lines monogenic for resistance derived from Kota were crossed and backcrossed to Little Club. Two additional lines with resistance from Kota, but which may carry more than one resistance gene, were crossed and backcrossed with Little Club in an attempt to isolate lines monogenic for resistance.

10. The Genetic Nature of Wheat Stem Rust Development on 'Hope' Wheat Has Been Clarified Insofar as the Host is Concerned. There are at least five genes in 'Hope' wheat which adversely affect stem rust development. How these correlate with the pathogen and the nature of the obvious interactions are still not defined. Chromosome 2B of wheat is extensively involved in reaction to the stem rust fungus. At least five genes occur on the chromosome and this indicates that search should be made for additional genes for resistance in the B genome of wheat and its relatives, especially Aegilops. (Cooperative with Missouri).

B. Diseases

1. Two New Chemicals Extend Range of Effectiveness for Rust and Loose Smut Control. Greenhouse evaluations of two new compounds (C735 and F461) obtained from the U.S. Rubber Company indicates that there is a potential for control of the wheat rusts by seed or soil treatment.

The compounds also reduce loose smut. In all tests at St. Paul, Minnesota, D735 was more effective and the dosage response slope was steeper than for F461. However, D735 has the lower water solubility, greater phytotoxicity, and poorer field performance. The latter result probably was a consequence of overdosage in field applications and consequent plant injury. The materials have additional qualities of interest in being effective with seed or soil treatment. Two weeks after planting both compounds reduced disease incidence from controlled inoculations by approximately 50%; F461 was still active against infection 4 weeks after planting. A formulation of nickel salt plus maneb gave the best overall field results in spray applications for rust control in Minnesota.

2. Five-year Old Rust Spores Able to Infect. In 1960, spores of Puccinia graminis tritici were stored in liquid nitrogen at Beltsville. Tests of these spores made in 1965 indicate a slight deterioration in viability, but no reduction in ability to infect wheat. No changes in pathogenic potential were detected. In Texas, nine among ten mycelial cultures of different fungi survived 3 to 8 weeks of storage in liquid nitrogen. Sclerotia survived in one case when mycelium did not.

3. International Rust Nursery Continues Effectiveness. A total of 1,776 wheats are currently under stem, leaf, and stripe rust tests at over 50 locations around the world. The stripe rust section was added this year to both the spring and winter wheat portions of the nurseries for the first time.

4. Rust Spores from Rain Related to Epidemics. Spores of both leaf and stem rust were collected in rain samples early in two growing seasons. Further observations indicated that the initial rust infections of susceptible wheats develop when spores are washed from the air by rain and fall on wheat plants. Detection of rust spores in early spring rain samples could help in forecasting epidemics of wheat rusts. In both years, spores of leaf and stem rusts were found in rain samples in early May when the first seedling leaves were the only receptive wheat foliage in Minnesota. Rust infection at this stage can have a significant impact on later rust development. If temperature and moisture conditions are favorable, the initial infection sporulates and produces a rapid increase in rust infection. Often, the amount of inoculum produced locally exceeds that carried in later in the growing season by winds from southern grain areas.

5. Dramatic Demonstration of Stem Rust Resistance. Resistance of leading hard red spring and durum wheats to prevalent races of Puccinia graminis provided adequate protection for the 1965 North Dakota wheat crop. The importance of stem rust resistance was dramatically demonstrated by the complete destruction of susceptible 'Langdon' durum grown in a 1 acre plot at Fargo. Langdon was effectively resistant five years ago.

6. Aerospore Sampler for Use in Aircraft. An aerospore sampler was designed and constructed in Kansas to answer the need of a quantitative sampler which could be operated in aircraft. The basic idea is to use the sampler to locate geographical centers of rust infection by sampling the air above and to study the effect of wind and air turbulence on the transport of rust spores. A prerequisite therefore is to be able to identify a given ground location with a given point on the trapping surface. The design utilizes 35 mm film, coated with a thin layer of silicone, drawn under an air intake orifice at a constant rate. Therefore, by knowing the speed of the aircraft and the speed of the film, geographical locations can be pinpointed on the film strip. The sampler was used on nine occasions during the spring and summer of 1965. Spore transport at 1500 to 2000 ft above ground appeared to be more vertical than horizontal and aerospore concentration was greater over geographical areas of high disease severity than over non-wheat areas or areas where rust severity was light.

7. Stem Rust Infection Affects Roots of Wheat. Heavy stem rust infection on adult wheat plants grown in nutrient solution caused an early and abrupt decline in root respiration followed by premature death of shoots in Minnesota tests. As suggested by similar earlier trials, reduced root function probably contributes to premature death of field-grown rusted wheat.

8. Cytokinins Alter Tissues Prior to Sporulation. Indirect evidence continues to indicate that cytokinins cause the characteristic alterations in mildewed and rusted wheat tissues that precede sporulation. Green islands with the associated accumulation of nitrogenous compounds produced by powdery mildew disease resembled tissues treated with synthetic cytokinin in senescing detached leaves. (Cooperative with Minnesota).

9. Haustoria Function Measured. Limited growth of an obligate parasite (Erysiphe graminis) was obtained after destruction of host cell protoplasts and after removal of haustoria (the parasite's feeding organ). Using special micro-culture techniques, fungus growth with haustoria present often were made to continue at near-normal rates for two days after host cells were torn open with micro-needles. With haustoria removed, growth continued at 1/5th normal rates for 2-3 days. Since growth occurred without direct contact between the fungus and intact host protoplasts, growth of this fungus must not depend on close coupling between metabolic systems of host and parasite. The micro-culture system offers an unusually promising system for elucidating mechanisms of obligate parasitism by rust and mildew fungi. (Cooperative with Minnesota).

10. Nonspecific Rust Resistance in Maize. The host-pathogen relationship of Zea mays and Puccinia graminis var. tritici was studied microscopically in fresh, cleared, and fixed tissues. Development of the fungus was the same on corn and wheat through appressorial formation. Four times as many penetrations occurred on wheat than on corn, and fungal growth in corn was interrupted in all stages of post-appressorial development. Fungal

growth ceased at or before contact with corn mesophyll cells, and with one exception, haustorial mother cells and haustoria were not formed.

Results of tests with genotypically diverse selections of host and fungus indicate that the resistance is nonspecific. The histological observations suggest that diffusates from P. graminis are deleterious to corn cells, and that inhibition of fungal growth in corn is due to the presence of a diffusable, toxic constituent. (Cooperative with Minnesota).

11. Black Point Investigated. Several fungi and one bacterium are implicated in black point, by recent culture tests in North Dakota and Texas and include species of Helminthosporium, Fusarium, Alternaria, Plenodomus, kantomonas, and others. A genetic basis is sought for differences noted among varieties of both durum and common wheat. Functional, morphological, and escape mechanisms are under study.

12. Photosynthesis Measured in Study of Septoria and Mildew Diseases. Growth processes in diseased tissues were evaluated by use of infra-red carbon dioxide analysis at Beltsville. When leaves are lightly infected with powdery mildew and pustules scattered or confined to leaf tips, a gradual decline in photosynthesis occurred with age of tissues, resembling senility. Heavy infections caused marked declines in photosynthesis. Septoria nodorum infections, even light infections, caused marked declines in photosynthesis with advancing age. Heavy infections drastically reduced photosynthesis 75% or more. Respiration and other responses are also under study.

13. Root and Foot Rot Research Expanded. In Texas, Sclerotium rolfsii was found to be pathogenic on wheat. Varietal differences were noted. In Washington, 100 clones of Fusarium, representing five species, were observed for pathogenicity. Field soil frequently contains 20-30 propagules/mm³ of soil virtually assuring that wheat plants cannot avoid contact with the fungus. Ascospores may represent a portion of the Fusarium inoculum in the field. Calonectria nivalis, perfect stage of Fusarium nivale and heretofore not reported from nature in North America, occurs on wheat in the fields in this region. Similarly, Gibberella roseum, perfect stage of F. roseum 'Avenaceum' and heretofore not reported from the Pacific Northwest, occurs on wheat in the region.

Studies on F. culmorum in the field have shown that, like other soil-borne Fusaria, it persists in the field as chlamydospores. Sometimes the number of propagules in the field ranges from 200-1,000/gm, depending on the field.

14. Cercospora Tolerance Levels. Work in Washington revealed a surprisingly high number of wheat selections with unexpected levels of tolerance to this pathogen which causes foot rot. A large percentage of selections with good tolerance in the foot-rot-inoculated plots during 1964 continued to have good tolerance in 1965. A few notable discrepancies, however, remain to be explained. Particularly encouraging are the unexpected accumulations of minor responses from crosses with parents assumed to be highly susceptible to foot rot.

15. Snow Mold Reactions Consistent. According to test results, Moro has a good tolerance to snow mold. Gaines and Burt are most susceptible. McCall, originally released for its known tolerance, continues to show it. Idaho and Washington collaborated in the development of selections from the World Collection that have good snow mold tolerance. Two of the best have been given C.I. numbers 14106 and 14107.

16. Correlation Demonstrated for Fungus Vector with Wheat Virus. A correlation has been demonstrated between the presence of Polymyxa graminis and the transmission of soil-borne wheat mosaic virus in Nebraska studies. In conjunction with previous results, this correlation indicates that the fungus is probably the vector of the virus. This is the first virus known to be transmitted by a fungus other than Olpidium. The exact relation of P. graminis to the virus and the mechanism of transmission must still be elucidated.

It appears that one obligate parasite, the virus, uses the transmission mechanism of another obligate parasite, the fungus, to carry it from host plant to host plant. In a way, the virus is a double parasite, depending for survival on two other organisms, which are in turn related by obligate parasitism.

17. Barley Yellow Dwarf Virus on Spring Wheat. Twenty-nine hard red spring wheat varieties infested in field plots showed an average reduction in yield of 35.3% with a range of 21.2 to 54.8% in South Dakota trials (Cooperative with Entomology). Number of kernels per head and weight of kernels accounted for most of the reduction but varieties did not respond equally to the virus for these characters. The number of heads per foot of row appeared to be little affected by the virus in this trial. BYDV produced large and dissimilar individual effects among 1800 entries, but the average effect was to delay heading 1.6 days and reduce height by 3.7 inches. Ten wheat entries from greenhouse screening trials for resistance to BYDV have been selected from 152 varieties tested in 1964 and 1965 as having some promise of tolerance or resistance.

Fourteen of more than 1800 wheats in a field trial for resistance to BYDV showed minimum symptoms in the field and were very promising in later greenhouse tests. These lines are being crossed with both winter and spring varieties.

18. Rust Development and IAA Associated. Characteristic derivatives of indoleacetic acid (IAA) were associated with rust development in Nebraska. Light stimulates IAA-oxidase activity. When plants are grown under incandescent light prior to inoculation, a reduction in infection was noted on susceptible plants and no infections on resistant plants.

19. Host-Parasite Rust Relationships. Ultrastructure studies being conducted through Duquesne University indicate that the flecking reaction noted in stem rust is not accompanied by establishment of the fungus even to a limited degree in the SR 6 line of wheat grown at 65°F in contrast to full invasion at 80°

20. Occurrence of Streak Mosaic in Ohio Wheat. Tests at Beltsville and Nebraska confirm that the 3A virus collected in Ohio is a strain or mixture of strains containing wheat streak mosaic virus. Wide-spread occurrence of this virus was observed in wheat and corn in Ohio. Serology, particle dimensions, blocking, and hosts attacked were among the lines of evidence accumulated.

21. Rust Races in Spain. Races of stem rust observed in Spain (P. L. 480 research) revealed three races that do not correspond to anything in the United States key. Likewise, four new races of leaf rust were identified. Seven wheat varieties including Timopheevi, Newthatch, N.D. 56-122, R.L. 2812 and R.L. 2813 were immune to stem rust when grown near infected barberries. Twenty varieties were very resistant.

C. Quality

1. High Protein Winter Wheats Promoted in Research Trials. Advanced high protein selections of Atlas 66 x Comance and Atlas 66 x Wichita were outstanding in performance in Nebraska. They combine such important characteristics as leaf and stem rust resistance, moderate winterhardiness, mid-season to early maturity, and high protein with good productiveness.

Amino acid studies of these high protein lines show that lysine contents for a given weight of protein are similar to their low protein parents. Therefore, higher protein contents offer a potential for increasing nutritional quality of wheat. (Cooperative with Utilization Division and University of Nebraska).

2. High Protein Spring Wheats. Justin and Lee, known high protein varieties, are exceeded significantly by new spring wheats.

Of several hundred foreign wheats tested in Montana, P.I. 170926 from Africa, Fronteira from Brazil, and P.I. 176217 from India, have some promise as high protein parental stock. These wheats appear to have a high yield potential as well as a high grain-protein potential.

3. Quality Unimpaired in Soft Wheat Reduced in Yield by Cereal Leaf Beetle. Damage from feeding of larvae of the cereal leaf beetle [*Oulema melanopa* (L.)] caused considerable losses in yield of grain from Monon wheat. No significant losses in protein content, pearling index, mill yield, and alkaline water retention capacity were noted from samples harvested in Michigan. Loss in yield of grain was brought about by reductions in kernel number and kernel weight and amounted to 21.3 g per 100 heads, or 23%. (Cooperative with Entomology Research Division).

4. Value of Small Kernels Negative in Flour Milling. Flours were milled from the following spring wheat samples: the unsized (control), the individual sized kernels (large, medium, small), and a sample with the small kernels removed.

According to milling data, the larger the kernels the greater the flour yield. Also, with the small kernels removed (approximately 5%), as much or more flour can be obtained as from the control. Cumulative ash curves showed that small kernels contribute high ash to the flour. Removing the small kernels produces no deleterious effects on mixing and bread baking.

5. Saturated Sterol Ester Determination for Species Identification. This test for semolina, farina, and mixtures of the two has been consistent in identification of durum from common spring wheat middlings. The method was extended to other classes in North Dakota studies.

Several samples of hard winter wheat were analyzed for saturated sterol ester content by thin-layer chromatography and densitometry. Variations in amount existed among the varieties, but all showed saturated sterol ester content greatly in excess of the amount present in durum semolina. Four samples of Italian soft wheat were analyzed, and two showed extremely low amounts of saturated sterol esters.

6. Pacific Northwest Flours Require More Chlorine to Adjust pH. Various bleaching procedures of two Pacific Northwest flours were compared with those of an unbleached cake flour and an unbleached cookie flour from Ohio, and one eastern soft wheat. The data show that more chlorine is needed for PNW flours and that the higher chlorine requirements are due to both a higher initial pH and a higher chlorine requirement to achieve the same amount of pH drop. Experience with a large number of PNW samples confirms this.

7. High Alpha Amylase Content Varies with Location and Variety. Over 250 samples were examined including 27 varieties grown in different locations in the Pacific Northwest from seven recent crops. The enzyme content varied over a wide range. Some samples of each variety had a high enzyme content but Baart, Ramona, Lemhi, Druchamp, Itana and Kharkof generally had high values. Location appeared to be a factor as the

driest locations produced grain with the lowest enzyme content. Neither nitrogen nor sulphur fertilizers appeared to influence the values obtained. The alpha amylase content can be high without obvious sprouting.

8. Bread Costs Increased by Excessive Mixing-Time Wheats. Typical mixograms of flours that vary in mixing requirement and tolerance show that mixing tolerance increases (slope of curve beyond the peak decreases) as mixing time increases to about four minutes under Kansas laboratory conditions.

Thereafter, mixing tolerance remains constant with increasing mixing time. Thus, selecting new progenies of wheat that have mixing requirements appreciably greater than about four minutes would not give increased mixing tolerance, and in many instances would result in increase of production costs.

9. Medium to Long Mixing-Time Buffers Frost and Weather Damage to Bread Wheat Quality. Wheat varieties that have Kaw-type properties are particularly desirable in areas where frosts or freezes are likely to occur during the boot, heading, or fruiting stages, in areas where climatological factors or irrigation practices usually contribute to abnormally long fruiting periods, and in the High Plains where temperatures above 90°F and low relative humidities are likely to occur during the fruiting period.

10. Particle Size Good Index of Hardness. Wheat moisture content was found to have a significant bearing on kernel hardness index values of eastern soft wheats. Increasing the moisture content results in high particle size index values, indicative of greater softness and finer granulation, but the effect of moisture on pearling index is opposite to that noted above. The anomalous behavior of wheat in the latter test is attributed to a toughening effect on bran by the added moisture, as well as to the greater relative importance of bran in the test. There is a parallel in trend between grain moisture effect on endosperm particle size and its effect on milling. This trend plus the fact that the entire kernel is tested leads one to believe that the particle size test is more useful than the pearling test in milling quality evaluation. At a given grain moisture content; however, both tests appear to appraise kernel hardness adequately.

11. Viscosity Values Adjusted to Uniform Protein Levels. Comparison of inherent protein quality in flours by the MacMichael viscosimeter method is sometimes not precise when large adjustments in viscosity values must be made for differences in protein content. A new relationship has been developed in Ohio between viscosity and protein content within a variety which appears to be valid theoretically and which fits the data. The third power of protein quantity (based on 3-dimensional

swelling in acid) has been found to be very highly associated with viscosity. By use of a computer, all adjustments have been calculated for the usually encountered range of values.

12. Wheat-Soya Flours. Carbon dioxide production in soya-wheat flour doughs was unaffected by the particle size of the soya flours, but was reduced by heat treatment or by desugaring the soya flour. Gas production was higher in doughs containing coarse soya particles than in finely pulverized soya-flour. Soya flour of intermediate size produced the most gas. Excessively toasted soya flour reduced gas retention more than did slightly toasted flour. Soya flour with a protein dispersibility index of about 40% gave best results. The studies indicated that lowered loaf volume of soya-containing doughs was, at least partly, explicable by impaired gas retention capacity in these Kansas trials.

13. Urea Extraction of Protein. Extraction tests in Kansas with 3M urea at 40°C yielded an average of 67.9% protein from wheat, 67.3% from wheat flour, 66.2% from rye, 71.2% from oats, 46.7% from barley, 32.1% from rice, 22.3% from corn, and 24.6% from Sorghum. Differences in dispersible proteins seem to be affected by the disaggregating and hydrogen-bond-disrupting effect of urea, and seem to be related to amino acid composition and functional properties of cereal proteins. The urea-dispersible protein fraction in wheat flour was higher than in whole wheat.

Tests revealed that dispersibility of wheat flour proteins in 3M urea is a sensitive parameter to evaluate the effects of heat treatment on drying damp preripe wheat and the effects of steam conditioning of wheat on bread-making potentialities of the flours. Dispersibility of proteins decreased with length and severity of heat treatment and paralleled damage to rheological properties and bread quality.

14. Amino Acid Composition of Proteins in Maturing Wheat. During maturation in Kansas the concentration in protein of lysine, aspartic acid, glycine, alanine, and valine decreased; concentrations of glutamic acid and proline increased substantially; and serine, leucine, tyrosine, and phenylalanine increased slightly. Flour from these hard red winter wheat samples have less lysine, histidine, arginine, aspartic acid, threonine, glycine, and alanine. Glutamic acid and proline are greater in flour than in wheat proteins.

15. Free Amino Acids in Maturing Wheat. The number and concentration of free amino acids decrease strikingly during maturation. Some could not be detected at all in fully ripened grain. Variety differences were not evident among hard red winter wheats observed.

16. Amino Acids and Protein Levels. The contents of almost all of the amino acids increased as the protein of the wheat from diverse sources and flour samples increased. Only cystine and methionine in the wheat, and lysine, cystine, and methionine in the flour failed to follow

this pattern. Milling of wheat to flour lowered (in decreasing order) concentrations of lysine, arginine, aspartic acid, glycine, alanine, tyrosine, histidine, threonine, and valine. The concentrations of glutamic acid, proline, and phenylalanine were higher in flour than in wheat. The concentrations in protein of basic amino acids, glutamic acid, and threonine were significantly correlated with certain rheological properties or bread-making potentialities of the flours. Proteins of hard red spring wheats contained less lysine, arginine, and methionine, and more cystine, than hard red winter wheats.

17. Lipids and Vegetable Shortening Effects on Flours. Bread was baked from flour milled in Kansas from hard red winter, hard red spring, soft red winter, durum, and club (white) wheat varieties, each from the 1963 and 1964 crop. Loaf volumes were increased 78 to 195 cc and crumb grains improved by adding 3 g vegetable shortening per 100 g of flour. The improving effect increased steeply from additions of up to 1.5 g shortening, and thereafter increased only slightly up to 4.6 g shortening. Adding 0.5 g lipids isolated from 6 flours to a composite hard red winter flour almost equaled the improving effect of 3 g shortening; adding 0.5 g nonpolar flour lipids had very little effect; and adding 0.5 g unfractionated, original flour lipids had an intermediate effect.

Durum wheat samples contained the most lipids and the highest concentration of nonpolar lipids. The bread-making wheat varieties had a lipid content which was consistent for the two years studied. Hard red spring wheat samples contained more total and nonpolar lipids than hard red winter wheats. The polar lipid content of wheats from the two classes was essentially equal. The wheats contained substantially more total lipids than the flours milled from the wheats. Nonpolar lipids constituted about one-half of the flour lipids and two-thirds of the wheat lipids.

In North Dakota, the pasting properties of durum and hard red spring wheat flours were compared after altering the lipid content. Nonpolar lipids, when removed, resulted in lower viscosity peaks. The primary effect was the prevention of hydration of the micellular regions of the starch granule. Polar lipids, in contrast, prevented hydration of the amorphous regions of the granule.

18. Monosomic 1-D Lines. Wheat-meal-fermentation-time values of monosomic 1-D chromosome lines deviate sharply from disomic materials. Electrophoresis, sephadex column, sulflaydral content, amino acid analysis, and serology so far fail to account for the observation (Montana Grant).

19. Starch Content of Bran, Meal, and Flour. Starch clinging to bran is an indication of poor milling properties. Starch in bran is now readily determined by modified flour-starch methods developed in Ohio involving enzymatic and polarimetric procedures. Correlation values by the two procedures was 0.99 on 127 comparisons.

D. Physiology and Culture

1. Roots Retain Small Amount of Nitrogen. Studies in Montana with five genotypes sampled nine times during the growing season indicate that roots constitute a small part of total plant growth and that the amount of nitrogen contained in roots would have little or no effect on grain protein even if all of it were translocated to the kernels. There is a tendency for varieties with greatest amounts of nitrogen in their top growth to have the greatest amounts of nitrogen in their kernels.

2. Leaf Metabolism Factor in Translocation of Nitrogen. Of four varieties grown in Nebraska in 1963 and 1964 and analyzed for foliage NO_3 content, Scout had the last amount throughout the sampling period in 1963; in 1964 consistent differences could not be detected. Relatively high foliage nitrogen of all varieties in 1963 was associated with high grain protein, whereas both foliage nitrogen and grain protein were lower in 1964. Field studies of the effect of defoliation on protein accumulation was continued. Early defoliation tends to reduce the nitrogen content of the vegetative portion of plants throughout the grain maturation period, but late defoliation has no consistent effect. Two selections of Atlas 66 x Comanche were consistently higher than other varieties in grain nitrogen at all stages of kernel development. Early defoliation of these selections markedly reduced the nitrogen content of their grain, whereas the reduction in lower protein varieties of comparable maturity was minor.

3. Peroxidase High in Dwarfs, Gibberellin-3 Effects. Chemical analyses have shown peroxidase content is nearly twice as great in dwarf plants as in normal plants when both are grown at 65°F. Dwarfs and normal plants grown at 85°F had about the same concentration of peroxidase. In Oregon trials, gibberellin-3 (GA-3) did not influence peroxidase concentration in normal and dwarf plants grown at either temperature.

Most dwarfs head and produce seed when the temperature is held at 80°F during the latter stages of growth. Two hybrids, Early Defiance x Kenya Farmer and Cedar x Kenya Farmer, are exceptions to this generalization. They did not head unless held at 80°F throughout the entire growth period. This need for maintaining an 80°F temperature throughout the entire growth cycle was circumvented by applying GA-3. A 5,000 ppm solution of GA-3 was applied as a spray to dwarf plants about 6 weeks old at the time temperature was raised from 70° to 80°F. Plants treated in this manner produced 25 to 100 seed each.

4. Vernalization Blocked by Growth Retardants. Four growth retardants were tested for their ability to block vernalization in Oregon trials. The vernalizing seedlings were treated for 5, 4, 3, 2 and 1 weeks with the test solutions. The growth retardants 2-chloroethyl-trimethyl-ammonium chloride (CCC), dimethyl hydrazide of succinic acid (B-995), and dimethyl-hydrazide of malic acid (C-011), all blocked the vernalization response in a cold-treated winter wheat. An antisteroid substance, 7997-A₃, did not block either the vernalization response of the winter wheat or the flowering response of the spring wheat.

5. Vernalization Products Resist Leaching. In Oregon, attempts were made to extract water soluble substances associated with the vernalization process. A small apparatus was constructed to permit controlled cold air and cold water to flow through a tube containing vernalizing wheat seedlings. At water flow rates up to 90m/hour/100 seedlings, substances required for the vernalization and subsequent flowering responses were not leached out of the young winter or spring wheat seedlings.

6. Bare-crown-freezing Technique Used on Wheat. A technique borrowed from oats is proving useful in Washington in evaluating wheat selections for cold hardiness. Field-grown seedlings are dug, tillers cut off at the soil line and crown roots removed. The trimmed crowns are sealed in a plastic bag, frozen, and allowed a standard recovery period. Tiller growth can then be related to cold hardiness.

7. Nutritional Requirements of Dwarf Bunt Fungus. Sucrose was the most effective carbohydrate in promoting mycelial growth. L-asparagine induced better growth than 7 other nitrogen-compounds. Thiamine was the only vitamin required and phosphorus, potassium, magnesium and sulphur were required elements. Calcium inhibited growth. A pH of 6.0 to 8.0 was optimum. (Cooperative with Oregon).

8. Sulphydryl and Disulfide Concentrations and Vernalization. Tests in Oregon showed that varieties differing in growth habit may differ in sulphydryl and disulfide values. These values, however, are not directly correlated with the vernalization process.

9. Root Systems and Lodging Relationships. In Israel (P. L. 480 research), it was found that the spreading angle of the roots is related to differences in "root lodging" of wheat varieties and that this characteristic is affected less by environment than by number of roots and number of tillers. Culm height, culm diameter, culm stiffness, etc., also may alter the manifestation of root anchorage of plants. The growth retardant, CCC, reduced general lodging and, in some trials, increased the grain yield by 30%. Whether root development is affected is being investigated.

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OAT AND BUCKWHEAT BREEDING, GENETICS, DISEASES, AND CULTURE
Crops Research Division, ARS

Problem. Most of the United States oat crop is produced under warm, humid conditions highly favorable for disease development. The major limiting factor for profitable oat production in the United States has been a constant acute need for new disease-resistant varieties. Other problems are lack of adequate straw strength, response to high fertility, adaptation, nutritive value, winter hardiness, yield, and grain and forage quality.

Buckwheat has been long neglected from the standpoint of breeding for increased yield, grain quality, straw strength, disease resistance, adaptation, etc.

USDA AND COOPERATIVE PROGRAM

The Department has a continuing long-term research program involving pathologists, geneticists, and agronomists engaged in both basic and applied research in an attempt to solve the problems besetting the oat crop. The ultimate objective is to stabilize production, increase efficiency, and improve the quality of oat grain and forage wherever it is grown. Most of the work on the breeding, genetics, and diseases of oats is conducted in cooperation with State Experiment Stations in Arizona, California, Florida, Missouri, New York, Pennsylvania, South Dakota, Texas, Virginia, Washington, and Wisconsin; the Puerto Rico Agricultural Experiment Station, and at Beltsville, Maryland. Oat research is being conducted under three P.L. 480 projects. One in Poland on crown rust of oats; a second in Colombia, S.A., on extremely virulent races of oat stem rust; and the third in Israel on collecting and screening wild species of oats for resistance to virulent races of stem and crown rust.

Buckwheat breeding and genetics are limited to an industry-supported graduate student at University Park, Pennsylvania.

The Federal scientific effort devoted to research in this area totals 13.3 professional man-years. Of this number, 5.6 is devoted to breeding and genetics, 7.5 to diseases, and .2 to physiology.

PROGRAM OF STATE EXPERIMENT STATIONS

A total of 37.4 professional man years is devoted to this area of research.

PROGRESS -- USDA AND COOPERATIVE PROGRAMS

A. Breeding and Genetics

1. Improved Oat Varieties. Bingham, a spring oat variety, was developed cooperatively by the Idaho Experiment Station and ARS. It has unusually

stiff straw and high yield potential and is outstanding when grown on irrigated land. Bingham is resistant to Victoria blight and race 8 of stem rust. The development of Bingham represents a significant step in the improvement of oats for Idaho and adjacent areas.

Jefferson is a dual purpose winter oat variety developed by the Georgia Agricultural Experiment Station and ARS. Grain and forage yields equal or surpass those of other commonly grown varieties in Georgia and adjacent States. It is suited to silage and grazing and produces an abundance of forage in late winter and early spring. Jefferson is resistant to Victoria blight, soil-borne mosaic, and many races of crown rust.

Tyler, a spring oat, is similar to the Tippecanoe variety, but distinguished by higher yield and exceptional standing ability. It is somewhat lower in test weight than Tippecanoe and is more susceptible to smut. Tyler was developed by the Purdue Agricultural Experiment Station and ARS.

Clintford, a spring oat variety, was developed by the Purdue Agricultural Experiment Station in cooperation with ARS. This rather short, new variety is outstanding in straw strength, yield, kernel size, test weight, and groat percentage. It possesses the "Landhafer" plus "Bond" types of resistance to crown rust, the D gene for stem rust, and is resistant to the Indiana races of the smuts. However, it is susceptible to some races of both rusts and smuts.

2. Irradiation of Oats. Irradiation induced genetic variability was cumulative in a recurrent irradiation study with oats in Iowa with the first cycle of irradiation having the greatest effect. The effect of irradiation upon variability is influenced somewhat by the oat variety and the character involved. The variation induced by irradiation is as heritable as that derived from hybridization.

Ploidy level in itself did not greatly influence the magnitude of genetic variability on types of mutation when diploid, tetraploid, and hexaploid oats were treated with thermal neutron irradiation. The magnitude of variation was influenced more by the character studied and the genotype treated. The types of mutation induced in the hexaploid are similar to those in the diploids and should be just as heritable.

3. Fostering Genetic Recombinations. A chemical mutagen (the herbicide Dalapon) and a composite of five monosomics were used in California to promote mass recombination within two oat populations. Seed lots of the M₂ and M₃ generations of the World Oat Collection composite treated with the mutagen are being distributed.

4. Hill Plots for Oat Testing. Approximately 300,000 hill plots have been used in Iowa. The genetic correlations between rod rows and hill plots for several agronomic characters have ranged from 0.96 to 0.98. Coefficients of variation for the components of yield were similar for rod

rows and hills, whereas the coefficient of variation for yield itself was 2 to 5 times larger for hills than for rod rows. Much of the experimentation which currently employs rod rows could be done in hill plots, but final evaluation should be done in rod rows.

5. A Technique for Determining Cold Resistance. A technique of freezing hardened winter oat plant crowns in moisture proof bags was developed to determine cold resistance. Readings can be made as early as 2 days after freezing. The correlations between survival in freezing tests and average field survival in uniform nurseries were highly significant and ranged from 0.716 to 0.883. This new technique should afford a rapid and accurate test for screening large populations of oat seedlings for different levels of cold resistance.

6. New Genes for Oat Improvement in Avena Sterilis. An extensive collection of the hexaploid Avena sterilis being obtained through a P.L. 480 project in Israel is yielding apparently new and different genes for resistance to virulent races of crown and stem rust, soil-borne mosaic, and the barley yellow dwarf virus. Unusually high grain protein content and significant variability for kernel size, vigor, maturity, and many other agronomic characters have been observed. The collections may well represent the most valuable reservoir of genes for oat improvement discovered to date. Fortunately, genes in A. sterilis can readily be transferred to cultivated oats. Numerous crosses have been made for inheritance studies and for transfer of the new genes to desirable agronomic types.

7. World Oat Collection. During 1965, the World Oat Collection received 102 selections of cultivated oats from 20 States and 3 foreign countries and 682 samples of Avena species from 7 countries. The Avena species included 630 collections of the hexaploid A. sterilis from a P.L. 480 project in Israel.

8. Tetraploid Buckwheat. An agronomically promising tetraploid type of buckwheat has been developed in cooperation with the Pennsylvania Agricultural Experiment Station. It is superior to the commonly grown diploid varieties for yield and lodging resistance. The tetraploid selection is being intensively tested and a preliminary increase obtained with the expectation that it will be named and released.

B. Diseases.

1. Cross Protection and Mutual Exclusion by Strains of BYDV. Seedling oat plants inoculated with a mild strain of barley yellow dwarf virus (BYDV) were protected against subsequent invasion by either of two severe strains of the virus under field conditions. When seedlings in the early one-leaf stage were simultaneously inoculated with the three strains, mutual exclusion was observed. Mild symptoms appeared soon after inoculation and were followed by complete recovery from symptoms and no recovery of the virus. When plants in a more advanced stage of growth were

similarly inoculated, the infection induced severe symptoms from which the plants never recovered. The three strains of BYDV appear to be closely related.

2. Loss of Vector Specificity Following Double Inoculation by Two Strains of BYDV. When oat seedlings were doubly inoculated with two vector specific strains of BYDV (one transmitted by Rhopalosiphum padi and the other by Macrosiphum avenae), infected plants produced symptoms more severe than those caused by either strain alone. In subsequent transmission from doubly infected plants, the vector specificity still prevailed in tests with M. avenae, but not in parallel tests with R. padi. When inocula from doubly infected plants were injected into aphids or when aphids fed through membranes on such inocula, the apparent breakdown of vector specificity occurred in some tests with R. padi, but not in parallel tests with M. avenae.

3. Comparison of Vector Specific Strains of BYDV. Vector specific strains of BYDV, transmitted by a single aphid species, generally cause less severe symptoms on oats than strains transmitted nonspecifically by several aphid species. Nonspecific strains transmitted by both R. padi and M. avenae have been most prevalent in Illinois where infected plants usually appeared first along the margins of the field and later often developed in circular spots. The disease has caused severe damage when infection occurred early in the season. In New York, where the vector specific strain transmitted by M. avenae has been most common, the first infected plants are usually distributed at random over the field, and losses have been generally less severe than in Illinois.

4. Resistance to Virulent Races of Crown and Stem Rust in Avena Sterilis from Israel. More than 100 entries possessing high seedling and adult resistance to the extremely virulent race 264 of oat crown rust have been found among collections of the hexaploid A. sterilis obtained through a P.L. 480 research project in Israel. Seedling resistance to race 264 has not been found among cultivated oats. The 264 crown rust resistance has been observed in greenhouse tests of seedling plants at Beltsville, Maryland, and Ames, Iowa, and in field tests of adult plants in a 264 nursery in Puerto Rico. Seedling resistance to crown rust race 326 was observed in 142 A. sterilis entries in Iowa, and adult resistance to a composite of races (including 326) was observed among 80 entries grown in Georgia, Florida, and Texas. Seedling resistance to race 6 group (6, 6A, 6AF) of stem rust was observed at Beltsville in 38 entries of the A. sterilis collections. The types of response observed in A. sterilis indicate the presence of a large number of different genes controlling resistance to crown and stem rust.

5. Effect of Crown Rust Measured by Kernel Density. Kernel density is a rather precise and readily obtainable measure of tolerance and resistance of oat varieties and selections to crown rust infection when the data are obtained under both rusted and nonrusted conditions. This measure can be

used instead of yield, test weight, and kernel weight in testing early generation material when seed is limited or when time and labor are limited.

6. Effect of Victorin on Krebs Cycle. Victorin, the causal agent of Victoria blight, caused increased production of malic acid through a CO₂ fixation reaction in susceptible oats. The increased citric acid metabolism was mediated primarily through the pyruvate - Co A system of malic acid which is released from the mitochondria and which acts as a buffering agent against excess cations that leak from the vacuole.

7. Effect of Light Intensity on "D" Genotype. Research conducted under a P.L. 480 project in Colombia and Iowa showed that 1, 2, 4, and 6 days of exposure of the "D" genotype to high light intensity (3,500 f.c.) would cause a breakdown in seedling resistance of plants held at 1,000 f.c. The critical period for effect of high light intensity was 9 to 12 days after inoculation.

C. Culture.

1. Factors Affecting the Survival of Winter Oats. Snow cover and relatively high soil moisture increase the chance for survival of winter oats. Survival was better on silt than sandy soil. Infection with soil-borne mosaic or barley yellow dwarf virus can materially reduce winter survival. Wintok has been the most winter hardy of all named oat varieties grown in the Uniform Winter Oat Hardiness Nurseries.

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RICE BREEDING, GENETICS, DISEASES, AND QUALITY
Crops Research Division, ARS

Problem. Rice in the United States is grown on a wide range of soil types and under quite different climatic conditions and cultural methods. Various types of rice are required to meet the demands of consumers and processors. The different environmental and climatic situations where rice is grown create a variety of contrasting problems in respect to disease and insect pests. The improvement in rotation systems, the increased use of nitrogen fertilizer, and better weed control methods have made necessary a modification of breeding objectives. Varieties are required that have seedling vigor under adverse conditions, such as low temperature of soil or water, so that good stands can be established in a short time in order that weed competition can be minimized. Herbicides are available for the control of many of the serious grasses and broadleaf weeds in rice. This enables the growing of short-season varieties because weed competition during the vegetative stage can be eliminated. High rates of N fertilizer increase the susceptibility of rice to blast, kernel smut, and lodging. Thus, the breeding of varieties resistant to these diseases and to lodging is of increasing importance. The nutritive value of rice also must be improved by increase of quantity and quality of protein. The exacting requirements of consumers require that all breeding lines be evaluated for milling, cooking, and quality characteristics.

USDA AND COOPERATIVE PROGRAM

The Department has a continuing long-term program involving plant breeders, geneticists, plant pathologists, and chemists who are engaged in basic and applied research to develop improved varieties and better methods for controlling diseases. This research is designed to stabilize production, increase the efficiency, and improve the quality of rice. Rice research is conducted at Beltsville, Maryland; and in cooperation with State Agricultural Experiment Stations at Stuttgart, Arkansas; Biggs, California; Baton Rouge and Crowley, Louisiana; and Beaumont, Texas. The work at Biggs, California, is also in cooperation with the California Cooperative Rice Research Foundation, Inc.; and at Beaumont, Texas, with the Texas Rice Improvement Association, and with private enterprises interested in rice quality.

A cooperative research project on the rice blast disease with the Japanese Ministry of Agriculture and Forestry, which is a part of the U.S.-Japan Scientific Cooperative Program, was continued throughout the year.

Rice research also is conducted under six P.L. 480 projects with (1) the Central Rice Research Institute, Cuttack, India, on the nature and variability of resistance of rice to the blast fungus and in physiological specialization in the causal organism; (2) the Institute of Botany, Academia Sinica, Taipei, Taiwan, on cytogenetics of rice and its related

species; (3) the Taiwan Agricultural Research Institute, Taipei, on physiological races of rice blast fungus; (4) the Taiwan Provincial Chung-Hsing University, Taichung, on genetic studies of mutations induced by radiation; (5) the Central Rice Research Institute, Cuttack, India, on chemical mutagenesis of rice; and (6) the India Agricultural Research Institute, New Delhi, India, on the collection and study of cultivated rices in northeast India and the assessment of the possibility for mutational rectification of undesirable trait prior to the use of wild rice in breeding.

The federal scientific effort devoted to research in this area totals 8 professional man-years, of which 5.0 is devoted to breeding and genetics; 2.0 to diseases; and 1.0 to quality.

PROGRAM OF STATE EXPERIMENT STATIONS

A total of 13.9 professional man-years is devoted to this area of research.

PROGRESS--USDA AND COOPERATIVE PROGRAMS

A. Breeding and Genetics

1. Improved Rice Varieties. 'Dawn' is a short season, long-grain, lodging and blast resistant variety developed cooperatively by the Texas Agricultural Experiment Station and ARS. Dawn is similar in grain type and quality to Bluebonnet 50 and Belle Patna and it is intermediate in maturity compared with these two varieties. The outstanding characteristic of Dawn is its resistance to the pathogenic races of Piricularia oryzae known to occur in the United States. Enough seed was released from the Arkansas, Louisiana, and Texas Rice Experiment Stations to plant about 1300 acres.

'Nova 66' is a short season, medium-grain, lodging and disease resistant variety developed cooperatively by the Arkansas Agricultural Experiment Station and ARS. Nova 66 is similar in maturity and grain type but it is more resistant to lodging and diseases compared with Nova and Nato. In an experiment at Stuttgart, Arkansas it was shown that with Nova 66 plant height was reduced 7 inches, lodging was decreased from 80 to 2 percent and yield was increased from 5063 to 7059 lb/A when 80 lb/A of nitrogen was applied 67 as compared to 43 days after emergence. These results point out the importance of applying nitrogen fertilizer as a topdressing at the proper time.

2. World Rice Collection. About 100 additional varieties from the World Collection of rice varieties were grown at Stuttgart, Arkansas and Beaumont, Texas. These varieties were classified for certain agronomic characters of the plants and physiochemical characters of grain. About 80 new introductions were received and grown at Beltsville, Maryland.

3. Genetic Diversity. An experiment was conducted at Crowley, Louisiana to compare the yield of mixtures with pure stands of the varieties in the mixtures. No yield advantage for the mixtures was shown.

4. Inheritance of Amylose Content. At Beaumont, Texas a study was made on the mode of inheritance of amylose content. This is an important factor as the cooking characteristics of rice is closely associated with the amylose content. In two crosses it was found that the varieties studied differed in one major gene and 2 or 3 modifying genes controlling amylose content.

5. Cytogenetic Studies of Rice and Its Related Species. This research was in Taiwan under a PL 480 research project. In the cross Oryzae sativa (Genome = AA) x O. officinalis (Genome = CC) the sterile hybrid was $2N - 24$ (AC). When the F_1 was backcrossed to O. sativa a triploid (Genome = AAC) was obtained. Similar results were observed in crosses of O. sativa with O. australiensis, O. brachyantha, and O. breviligulata.

6. Genetic Studies of Characters Induced by Irradiation. Under a PL 480 research project in Taiwan, studies showed that length of internode, panicle weight, plant height, and degree of extrusion of panicle from sheath are negatively correlated with the degree of lodging.

B. Diseases

1. Blast. Research on a joint project conducted at Beltsville, Maryland and Beaumont, Texas, in cooperation with Japan, to establish a set of varieties for differentiating pathogenic races of Piricularia oryzae and devise a system for numbering these races was completed. Eight varieties were chosen as differentials. On the basis of the results of the study, 32 international races were recognized in eight race groups. The international races are prefixed by the letter "I", followed by the letter A, B, C, D, E, F, G, or H which shows the race group. Individual races are designated by an arabic numeral that follows the group letter designation. The differential varieties are Raminad str. 3, Zenith, NP-125, Usen, Dular, Kanto 51, Sha-tiao-tsao (S) (C.I. 8970), and Caloro.

Under a PL 480 research project in India 14 races of Piricularia oryzae could be identified from a group of 17 isolates with 12 varieties used in preliminary trials under the U.S.-Japan blast study. In India, 35 pathogenic races of P. oryzae have been identified.

Under a PL 480 research project in Taiwan, results show that pathogenic races of Piricularia oryzae occur in Taiwan that are not the same as races that have been found in the United States. Resistance to these races is available for use in developing resistant varieties.

2. Seed Rot. Research on methods to control "seed rot" in water-seeded rice and "damping-off" in drill-seeded rice were conducted at Baton Rouge, and Crowley, Louisiana. A laboratory method for evaluating the effectiveness of chemicals used for treating seed was developed. This method gave results that were in agreement with results from field experiments. The laboratory test, which simulates water seeding in the field, is a practical way of screening new chemicals before field testing.

3. Hoja Blanca. Research was conducted at Baton Rouge, Louisiana on the effect of rice plant age on reaction to hoja blanca virus. Plants of 'Arkrose' and 'Gulfrose' when inoculated in the first leaf stage were susceptible, whereas, plants of these varieties when inoculated in the second or third leaf stage were resistant to hoja blanca virus.

C. Quality

Classification was made of the World Collection for quality characteristics that are associated with cooking and processing or nutritional level. The characters studied were grain size and shape, bran color, amylose content, starch-iodine-blue value, alkali value, protein content, and parboiling-canning stability. The range of values for these characteristics showed varieties quite different from present U.S. rice varieties. Some varieties for example were distinctly higher in protein than commonly grown U.S. varieties. This information is being used in breeding programs.

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ALFALFA BREEDING AND GENETICS, DISEASES,
PHYSIOLOGY AND CULTURE
Crops Research Division, ARS

Problem: Alfalfa in the United States is grown on about 30 million acres under a wide range of environmental conditions. The crop is subjected to many pests and climatic hazards. Resistant and adapted varieties are needed in each major producing area to cope with existing production problems, stabilize production, and increase efficiency of production. Research should be strengthened especially on developing multiple pest resistance and improving quality. Some problems are not readily solved by breeding, and their solution depends on management or other measures. Practical solutions rely on a continuing program of basic research.

USDA AND COOPERATIVE PROGRAM

The research program includes: Basic breeding and genetic studies; breeding for resistance to alfalfa weevil, potato leafhopper, spotted alfalfa aphid, pea aphid, bacterial wilt, common leafspot, Leptosphaerulina leafspot and Cercospora leafspot; basic studies on pathogens causing disease; and cultural and physiological studies on harvesting, diseases, and temperature stress. Research is conducted at: University Park, Pa., Beltsville, Md., Raleigh, N. C., St. Paul, Minn., Brookings, S. D., Lincoln, Nebr., Manhattan, Kans., Logan, Utah, Reno, Nev., and Stoneville, Miss. Research on mechanisms of resistance to aphids and stem nematodes was initiated through cooperative agreements at Reno, Nev., and Raleigh, N. C., respectively. Work is in cooperation with the respective State Agricultural Experiment Stations, except at Beltsville, Md. Most entomological research is in cooperation with the Entomology Research Division.

There is a P.L. 480 contract at Perugia, Italy, to develop lines of alfalfa resistant to Verticillium wilt.

The Federal scientific effort devoted to research in this area totals 16.1 professional man-years. Of this number, 9.7 are devoted to breeding and genetics; 3.3 to diseases; and 3.1 to physiology and culture.

PROGRAM OF STATE EXPERIMENT STATIONS

A total of 44.1 professional man-years is devoted to this area of research.

PROGRESS -- USDA AND COOPERATIVE PROGRAMS

A. Breeding and Genetics

1. Improved techniques hasten progress on breeding weevil-resistant alfalfa. By improving techniques and combining several methods of screening, over 600,000 plants were evaluated in Beltsville laboratories for resistance to

the alfalfa weevil. To test the same number of plants in the field would have been prohibitive. Resistance tests can now be conducted throughout the year and with greater precision. Four successive tests are used to isolate resistant plants: 1. Seedlings are grown in growth chambers under high light intensities to reduce hypocotyl elongation, and infested with adult weevils at 12 days of age. When all but 1 or 2 percent of the seedlings have been eaten, weevils are removed and the remaining seedlings saved for further testing. 2. Leaf discs from plants selected in step 1 are tested individually with adult weevils to identify plants which deter adult feeding. 3. Plants selected from step 2 are tested for larval growth and survival. Individual plants are confined in a plastic cylinder and infested with 10 freshly hatched larvae. Eight days later, larval weight and survival are determined. 4. Plants selected from steps 2 and 3 are individually tested for oviposition differences. At Raleigh, N. C., plants resistant to oviposition preference were isolated.

2. Washoe alfalfa, a variety with multiple pest resistance released. Washoe was developed cooperatively with the Nevada Agricultural Experiment Station and released jointly with the Experiment Stations in Nevada, Arizona, California, Idaho and Oregon. The new variety is resistant to pea aphids, spotted alfalfa aphids, stem nematodes and bacterial wilt, and is best suited to irrigated areas of the Pacific Coast and inter-mountain regions where stem nematodes and pea aphids reduce yields of other varieties. Washoe is winter dormant with winter hardiness similar to Lahontan. Progress was also made in development of pest-resistant germplasm with other types of adaptation at Lincoln, Nebr., Logan, Utah, Manhattan, Kans., Raleigh, N. C., and Beltsville, Md.

3. Release of Delta alfalfa. A new variety named Delta was released in cooperation with the Mississippi Agricultural Experiment Station. In plantings on heavy clay soil in the Yazoo-Mississippi Delta area, Delta remained productive for 5 years. This appears to be due in part to its tolerance to root and crown rots, leafhopper yellowing, and certain leafspot diseases. The new variety was equal to or slightly better than standard varieties in hay yield.

4. Germplasm released to alfalfa breeders. MSA-C4 and MSB-C4, two unrelated, broad-based pools of alfalfa germplasm were released to breeders. They had undergone 14 cycles of recurrent selection at Raleigh, N. C., Beltsville, Md., or University Park, Pa., for pest resistance and general adaptability. They are vigorous and dark green in color, resistant to common leafspot, potato leafhopper yellowing and rust, and more persistent than commercial varieties in eastern tests. Variability for growth habit, time of flowering, and other agronomic characters were maintained during selection to enhance their breeding value.

5. Attached vs. excised leaflets for evaluation of insect resistance. At Manhattan, Kans., adult and nymphal survival of the spotted alfalfa aphid was higher on excised than intact alfalfa leaflets. In all cases where differences among clones were significant, survival on excised trifoliolates exceeded that on the intact trifoliolates of the same clones. Using the excised-trifoliolate procedure in resistance screening would generally underestimate the level of plant resistance.

6. Meiotic drive in diploid *Medicago sativa*. An apparent case of meiotic drive, a force capable of altering equilibrium frequencies in a population, was found in diploid alfalfa at St. Paul, Minn. A plant, designated as G-29, has one large and one normal satellite. Such a large duplication would normally be transmitted through the gametophytic screen at a significantly lower rate than the normal chromosome. However, when G-29 was crossed with plants carrying two normal satellites, all the progeny had one large and one normal satellite. The normal satellite was not transmitted at all, suggesting some meiotic mechanism which insured that all functional gametes carried the large satellite. In alfalfa, crosses of diploid females x tetraploid male usually produce tetraploid offspring through restitution of two nuclei after meiosis, producing 2x eggs. When G-29 is crossed with tetraploid alfalfa, gametes with two large satellites function more often than gametes containing only one large satellite. No gametes functioned which contained only normal-sized satellites.

7. Effect of preferential pairing in autotetraploid mating systems. At University Park, Pa., the theoretical effects of different degrees of preferential chromosome pairing in an autotetraploid population were determined. Of particular concern were effects at different gene frequencies on elimination and fixation of gametes and on homozygosity.

8. Inheritance of flower color. Published and unpublished data from diploid and tetraploid alfalfa flower color studies were summarized at Beltsville, Md., to consolidate all data on flower color inheritance into a single source, compare similarity of data among studies, set forth a uniform system of gene designations, compare interpretations from diploid and tetraploid studies, and reevaluate mechanisms of inheritance. This information was made available in bulletin form and will be useful in genetic and breeding studies where markers are needed.

9. Selection for resistance to foliar diseases reduces coumestrol content. Biochemical studies at Brookings, S. D., showed that coumestrol content of alfalfa in each of two broad germplasm pools decreased as a result of recurrent selection for resistance to each of two foliar diseases, common leafspot and rust. Coumestrol content was positively associated with resistance and with size and number of leafspot or rust lesions except for rust in the most susceptible class. Urediospores were high in coumestrol, which suggested that loss of spores following profuse sporulation lowered coumestrol content of very susceptible plants. Selection in alfalfa for

resistance to common leafspot and rust can be expected to lower coumestrol content of alfalfa grown under conditions favorable for development of these diseases.

10. Pest resistance improves forage quality. At Lincoln, Nebr., protein and carotene contents of experimental entries with resistance to leafhopper and foliar diseases were superior to the check varieties, Buffalo and Ranger, in the second and third cuttings. The greatest increases in quality were obtained in a 1965 seeding where the carotene content of N.S. 16 was 59 percent higher, and the protein content of N.S. 37 was 38 percent higher than Ranger.

11. Effect of inbreeding on yield. At Raleigh, N. C., there was a linear relationship between yield and the coefficient of inbreeding among experimental strains differing only slightly for coefficients of inbreeding. The results indicated an extreme sensitivity of alfalfa to very low levels of inbreeding, possibly due to the rapid loss of potential allelic interactions from trigenic and tetragenic loci.

12. Selection for resistance to Verticillium wilt. In Italy, plants which appeared resistant to Verticillium wilt were selfed and/or intercrossed for potential use in the United States.

B. Diseases

1. Root rot problem identified in Minnesota. A root rot caused by Phytophthora megasperma Drechs. was identified for the first time in Minnesota from three fields in widely separated areas. Nine of 11 Phytophthora isolates from these fields were highly pathogenic on seedlings and 3-month-old alfalfa plants tested in a greenhouse.

2. Mechanisms of resistance. Techniques were developed to determine how alfalfa resists disease and insect attack. At Raleigh, N. C., one of the techniques for stem nematode studies utilized a single nematode mounted in a drop of agar which could be observed microscopically for behavior in contact with a seedling. This technique is being used to study attraction and repulsion of nematodes by plants and mechanisms by which nematodes enter plants. At Reno, Nev., research on mechanisms of resistance concerned biochemical differences in alfalfa which might affect aphid growth and reproduction.

3. Foliar diseases reduce forage yield, protein, and carotene contents. In Kansas, controlling alfalfa leaf and stem diseases in irrigated field plots by weekly sprays of Dithane M-45, increased annual forage yield 18 percent and carotene content 21 percent, and decreased defoliation 18 percent. Protein percentage was not increased significantly, but total protein yield increased with forage yield. Annual forage yields in tons per acre for sprayed and unsprayed plots, respectively, were Cody 8.7 and 8.0, Lahontan 8.9 and 7.3, Ranger 8.5 and 7.2, and Vernal 6.6 and 5.2.

Control of Ascochyta and Leptosphaerulina diseases increased yields of Cody 4 percent, Lahontan 14 percent, Ranger and Vernal 19 percent each. Control of Cercospora increased yields of Cody 16 percent, Ranger 29 percent, Lahontan 30 percent and Vernal 38 percent. Corresponding increases in carotene were 17 percent, 20 percent, 45 percent, and 10 percent, respectively.

C. Physiology and Culture

1. Management of alfalfa for production of alfalfa meal. Large plots were commercially chopped and dehydrated in Nebraska to study management practices. Two-years' data showed that maximum annual yields of dry matter, protein, and carotene were obtained when alfalfa was cut always at 1/10-bloom stage. A comparison of three management systems on full-bloom alfalfa showed that multiple cutting, in which the top and bottom halves of the plant were harvested and processed separately, effectively separated forage into high and low quality products. Partial cutting of full-bloom alfalfa, in which the top half was harvested leaving the bottom half for the second cutting, harvesting the entire plant at pre-bud stage in the second cutting and at full-bloom in other cuttings, improved total season quality over a uniformly late cut treatment. Multiple cutting and partial cutting systems can be used without additional capital investment.

2. Biosynthesis of disease-induced coumestrol. Research at Raleigh, N.C., showed that the period of rapid accumulation of coumestrol in alfalfa leaves inoculated with Ascochyta leafspot or rust corresponded with the time of appearance of visible symptoms. C¹⁴ was incorporated into certain fluorescent compounds formed during disease development. Incorporation into the pathogen-induced coumestrol is under study.

3. Variety affects saponin characteristics of alfalfa. Research on saponins at Logan, Utah, provided additional information on differences among alfalfa varieties with regard to biological effects on other organisms. Bands of saponin separated by the TLC method had different effects on growth of the test organism Trichoderma, and these effects differed according to variety. The results indicated that reducing or modifying the saponin content of alfalfa by breeding and selection to improve nutritional value is a realistic goal.

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CLOVER AND OTHER LEGUMES BREEDING AND GENETICS, DISEASES,
QUALITY AND VARIETY EVALUATION, AND PHYSIOLOGY
Crops Research Division, ARS

Problem. At least 50 legume species are either of regional or national importance or are potentially valuable for forage, provided adapted palatable varieties can be developed. One or more of these are grown on most farms or ranches for pasture, hay, silage, soil conservation, or multiple uses. Lack of adaptation to adverse climatic conditions, coupled with losses due to diseases and insects, can reduce farm value of these legumes by 50%. All *Trifolium* species of current economic importance lack persistence and resistance to specific diseases and insects. Inter-specific hybridization, possibly the key to further plant improvement in *Trifoliums*, has been extremely difficult to date. Sweetclover improvement needs include low coumarin, large seed, and resistance to sweetclover aphid and weevil. Trefoil needs improvement with respect to seedling vigor, resistance to root rots, broader adaptation, and greater productivity as a grazing plant on soils having poor internal drainage. More nutritious (low tannin) and more productive varieties of *sericea lespedeza* are needed for eroded acid soils of the South. Lupines, to be a reliable crop for winter grazing, must be more winterhardy, low in alkaloids, and resistant to diseases and insects. Greater mid- and late-summer production in permanent pastures is needed to improve crownvetch--a long-lived, strongly creeping-rooted legume widely used for erosion control.

USDA AND COOPERATIVE PROGRAM

The Department has a long-time basic and applied research program under way largely in cooperation with State Agricultural Experiment Stations. Research involves breeding, pathological, or physiological studies on red clover at Madison, Wis., University Park, Pa., and Lexington, Ky.; sweetclover at Lincoln, Nebr.; white clover at Clemson, S.C.; crimson clover at State College, Miss.; other perennial clovers at Ft. Collins, Colo.; birdsfoot trefoil at Ithaca, N.Y., Columbia, Mo., Blacksburg, Va., Stoneville, Miss., and Tifton, Ga.; lupines and tick clover at Tifton, Ga.; *lespedeza* at Raleigh, N.C., and crownvetch at Raleigh, N.C., Columbia, Mo., and Blacksburg, Va. In addition to the cooperative program, basic and applied research involving breeding and/or disease investigations are under way on red clover, birdsfoot trefoil, and crownvetch at Beltsville, Md. Most entomological research is in cooperation with the Entomology Research Division.

Improvement of winter annual clovers in addition to crimson clover was initiated at State College, Miss. Pathological studies on white clover at Durham, N.H., were terminated. Contract research of 3 year's duration, on nature of resistance to sweetclover weevil, was initiated jointly with the Entomology Research Division at Lincoln, Nebr.

Contracts with appropriate research departments in foreign countries, supported in part by PL 480 funds, involve research on: (1) Metabolism of alkaloids in legumes and their physiological role, Poland; (2) Forms of nitrogen in legumes, Poland; and (3) Collection and evaluation of native and introduced legumes, Brazil. A PL 480 contract on population studies and selection in berseem clover and the closely related taxa was initiated in Israel.

The Federal scientific effort devoted to this research totals approximately 16.2 professional man-years, with 10.1 devoted to breeding and genetics, 2.7 to diseases, 1.0 to quality and variety evaluation, and 2.4 to physiology.

PROGRAM OF STATE EXPERIMENT STATIONS

A total of 41.1 professional man years is devoted to this area of research.

PROGRESS -- USDA AND COOPERATIVE PROGRAMS

A. Breeding and Genetics

1. Red clover. Screening of lines and synthesis of varieties resistant to mildew, northern anthracnose, and three virus diseases was continued at Madison, Wis. Two recessive genes, producing absence of leaf markings as either homozygote or heterozygote, each produced a different leaf marking when crossed to other previously identified leaf marking genes. In Pennsylvania, field selections for resistance to internal breakdown (IB) had 50% less IB than did nonselected plants with comparable root size. Resistance to northern root knot nematode was found in both Pennscott and IB-resistant selections. All available plant introductions were screened at Beltsville for adaptation and for flowering type.
2. Sweetclover. Selection, progeny testing, and synthesis of varieties were continued at Nebraska for low coumarin, aphid resistance, and large seed. Lines selected previously for low coumarin were especially susceptible to attack by potato leafhopper. Selection for resistance to herbicide spray-drift was initiated within best commercial varieties. Tachinid flies were proven pollinators of sweetclover. Translocation across the graft union of the sweetclover weevil resistance factor of M. infesta was minimal; grafting did not increase crossability of M. infesta with any of four closely related species. An intermediate level of resistance to sweetclover weevil was discovered within M. dentata. Synthesis of o-hydroxycinnamic acid occurred in the above-ground parts of sweetclover plants with some translocation to roots; genotypes needed to determine the degree of dominance of the Cu gene over its recessive allele were obtained and vegetatively increased.
3. Crimson clover. F_2 populations to determine inheritance of six previously unreported qualitative characters in crimson clover were produced in

Mississippi. High temperature dormancy, effectively broken by transferring seed from high to low temperature, was found in inbred lines and polycrosses. S₅ lines within Dixie were especially desirable agronomically. Seven new synthetics were formulated. Relative performance of polycrosses was similar for seeded rows or broadcast plots; spaced plants were least reliable as a measure of polycross performance.

4. White clover. "Tillman" white clover was released cooperatively with the South Carolina Agricultural Experiment Station as superior to ladino in persistence and forage production. Screening for resistance to root-knot nematode and determination of level and inheritance of such resistance continued in South Carolina. Colchicine and embryo culture techniques were perfected for future interspecific hybridizations of white clover.

5. Perennial clover. In Colorado, a genetic factor, nonallelic to the S locus and interacting with a S allele to change the compatibility reaction of pollen from self-incompatibility to self-compatibility, was discovered in diploid alsike clover. The inheritance of a self-compatibility response to high temperature was found to be controlled by a single gene nonallelic to the S locus, with sensitivity to high temperature dominant in tetraploid alsike clover. All 42 noninbred zigzag clover plants studied were highly self-incompatible, but cross-compatibility studies yielded continuous variation for seed set and no evidence of intrasterile, interfertile groupings.

6. Birdsfoot trefoil. Foundation seed of Dawn, developed in the cooperative program at Columbia, Mo., was released to growers in 1966. Dawn is a long-lived pasture type, more resistant to root rot than Empire. New synthetics produced in cooperative programs at Ithaca, N.Y., and Blacksburg, Va., continue to look promising and are being regionally tested. Progress is being made through recurrent phenotypic selection at the above locations and at Beltsville, Md. Methods of strain synthesis are being studied in the cooperative program at Ithaca, N.Y. Four HCN negative (recessive) and two HCN positive clones produced seed of single crosses, double crosses, and four clone synthetics. Self and/or sib progenies were identified from HCN negative clones or single crosses. In spaced plants and field plots, more crossed plants survived than self or sib progenies. Four clone synthetics yielded more than double crosses due to the higher proportion of lower yielding sib progenies in the double cross. It would be possible to predict double cross performance on the basis of nonparental single crosses only when complete crossing occurs in both the double and single cross.

At Ithaca, N.Y., on well-drained soil, the yield of Narragansett alfalfa exceeded that of Viking trefoil by 11%, 4.44 tons vs. 4.01, respectively. On an adjacent poorly drained soil, trefoil produced 3.37 tons as compared to 1.84 tons for alfalfa--an increase of 83% in favor of birdsfoot trefoil.

7. Lupines. Rancher, developed in the cooperative program at Tifton, Ga., was released to growers in the fall of 1965. This is the first forage variety of blue lupines that carries resistance to anthracnose and gray leafspot and is genetically marked for ease of identification. All present

lupine varieties, including Rancher, are susceptible to winter injury at temperatures below 20°F. Greater winterhardiness has been found in an introduction from Portugal, and appropriate crosses have been made with desirable disease-resistant forage types. A number of F₄ and F₅ lines carrying winterhardiness, sweetness, and disease resistance survived at more northern locations in the South this past winter. Addition of winterhardiness to lupines will greatly increase the reliability and use of blue lupines for forage in the Southeast. A germplasm source of seed shatter resistance has been obtained from Australia which is being incorporated into winter-hardy forage types.

8. Crownvetch. Progenies of crosses of 20 widely different plants are being evaluated for differences in combining ability and as related to certain morphological characters in the cooperative program at Raleigh, N.C. In cooperative programs at Blacksburg, Va., and Columbia, Mo., studies are under way to secure long-lived, productive plants from available germplasm. Progenies of numerous selected clones are being evaluated at Beltsville, Md., under a system of frequent but partial defoliation to find productive types that will persist in permanent pastures under continuous grazing. Crownvetch appears to be resistant to many of the insects (meadow spittlebug, alfalfa weevil, etc.) that infest stands of commonly used legumes. The species is slow to establish and appears to be a better plant for grazing than for hay.

9. Sericea lespedeza. Breeding to improve the productivity of low-tannin lines is continuing in the cooperative program at Raleigh, N.C. Digestibility values for common and low-tannin-containing strains of sericea (in vitro technique) were 29.2 and 44.7%, respectively. Actual tannin values were 13.5 for the common or high-tannin strains and 3.2% for four low-tannin experimental lines. Backcrossing low-tannin plants to more productive types is being employed to secure more productive low-tannin varieties.

10. Desmodium and other summer-growing legumes. Desmodium uncinatum, a perennial forage legume, continues to look promising in cooperative studies at Tifton, Ga. Productivity of the species is being determined in association with other grasses and in pure stands. Lotononis bainseii, while looking good in fall and early spring, proved to be very susceptible to Rhizoctonia solani during summer. Siratro, Phaseolus atropurpureus, winter-killed. Strains of Dolichos lab lab and Glycine javanica are being evaluated in an effort to find a better summer forage legume for the Southeast.

11. Korean lespedeza. Yadkin, Korean lespedeza, developed in the cooperative program at Raleigh, N.C., has been released to growers. Yadkin is higher yielding in seed and forage and later in maturity than Korean. It carries resistance to tar spot and root-knot nematodes.

B. Diseases

1. Chemical nature of red clover crown pith susceptible to internal breakdown. Chemical analyses in Pennsylvania indicated no significant differences

in amino acids or in inorganic acids between healthy and diseased pith tissue; thin layer and paper chromatography revealed highly fluorescent spots in IB tissue that were absent in healthy tissue; the compounds were identified tentatively as phenols.

2. Roles of insects and diseases in red clover. Insecticides and/or cage growth improved plant stands and yields of red clover through two years of a study; fungicides did not. Effective treatments reduced incidence of viruses and controlled root-feeding insects. Cortical root rot was greater with inoculations of both Fusarium spp. and bean yellow mosaic virus than with either fungus or virus alone.

3. Effects of virus in birdsfoot trefoil. At Beltsville, Md., both tomato and tobacco ringspot viruses have been found infecting plants of trefoil. Comparative studies of the effect of TRSV on three clones of trefoil show infected plants to be significantly reduced in number and length of roots and in total production. In a limited study, 2.5% of the seed produced on TRSV-infected plants transmit the virus.

4. Factors affecting virulence of root-rotting fungi in birdsfoot trefoil. In cooperative studies at Columbia, Mo., a state-wide survey showed Leptodiscus terrestris, Rhizoctonia solani, Macrophomina phaseoli, and various Fusarium species to be associated with diseased roots. L. terrestris was found to be a destructive pathogen, but isolates varied in pathogenicity when tested on individual clones. At Beltsville, Md., pathogenicity of two species of Leptodiscus varied with temperature. Pathogenicity was enhanced at a soil temperature of 80° to 85°F. Culture experiments with the two species indicated that sclerotia are formed at a pH between 4.0 and 5.5. Sclerotia were not formed readily in a media above pH 6.0.

5. Longevity of cucumber mosaic virus (CMV) in lupine seed. At Tifton, Ga., seed from 108 diseased mother plants was stored 32 months. Approximately 11.5% had CMV symptoms indicating that infected seed may serve as a virus source for as long as 32 months.

C. Quality and Variety Evaluation

1. Clovers. Uniform variety tests of sweetclover, cooperatively conducted at nine locations in the United States and Canada in 1965, indicate that Evergreen, Denta, Goldtop, and Spanish were superior yielding sweetclover varieties from 1958 through 1965. In Nebraska, tests of red clovers from 1960 to 1965, Kentucky Syn A-1, Lakeland, Dollard, and Nebraska Common had superior ability to survive into the third year. Tillman was superior to all white clovers in stand at the end of a third year when tested in South Carolina in combination with tall fescue under rotational grazing management. Several new synthetic varieties of red clover were superior for forage yields and persistence in Wisconsin evaluations. In Mississippi, reseeds of Autauga and Chief crimson clovers were superior to that of Frontier crimson or Yuchi arrowleaf clovers.

2. Promising new legumes. Over 800 native legumes have been collected in Brazil and are being evaluated along with over 300 introduced species under a PL 480 project in Sao Paulo, Brazil. As a result of the preliminary evaluation, 11 seedstocks of the following genera, Calopogonium, Centrosema, Glycine, Lotononis, Phaseolus, Pueraria, Stizolobium, Stylosanthes, and Teramnus are being increased for more intensive evaluation studies.

D. Physiology

1. In vitro foam production and animal-bloating capacity of legume species. Studies in Kentucky indicated that all legumes tested contain proteins which under varying experimental conditions in vitro may produce equal volumes of foam. Foam production, as it relates to incidence of bloat in animals, is limited in nonbloating legumes by the tannin and pH of the rumen fluids.

2. Persistence of white clover improved by partial shade. In South Carolina, white clover plants produced greatest numbers of nodes, highest frequency of active axillary buds, highest frequency of axillary buds developing into stolons, and greatest number of rooting nodes when grown under plastic screen creating 33 percent shade. Results are interpreted in terms of reduced soil temperatures and increased moisture levels rather than reduction in light.

3. Seedling and axillary bud development in white clover as affected by temperatures. Earliest stages of germination were more rapid at 40° than at 16, 24, or 32°C, according to research at Clemson, S.C. Axillary buds were initiated in the axil of the first leaf by 13 days after start of germination at 16 and 24°. By 24 days, axillary buds were highly developed in seedlings at lower temperatures but not at 40°C. Stolon production was most vigorous at 24°C as early as the 6th week. The apical bud and its role in forming leaf primordia, the differentiation of the vascular system, and the development of cell types and tissues were followed histologically.

4. In vitro red clover pollen germination and pollen tube growth. At Lexington, Ky., an artificial medium was developed that supports red clover pollen germination and pollen tube elongation. The basic medium contains 50, 2,000, and 200 PPM of boric acid, calcium nitrate, and yeast extract, respectively; sucrose was also required; for optimum germination, 40% sucrose was essential; for pollen tube elongation, 25% sucrose was optimum. Optimum temperature for both germination and elongation was 18° and 25°C.

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FORAGE GRASS AND TURF BREEDING AND GENETICS, DISEASES,
QUALITY AND VARIETY EVALUATION, CULTURE AND PHYSIOLOGY

Crops Research Division, ARS

Problem. Some 90 introduced and native grasses are of agricultural importance in the United States for forage, turf, and soil conservation purposes. Of these, only 25 are included in active research programs. Up to 50 per cent of new seedlings are failures or near failures; and this staggering loss could be reduced materially with varieties possessing improved seedling vigor, seedling drought tolerance, and disease resistance. Diseases reduce the yield and quality of forage and seed and contribute to the loss of established plantings grown for either forage or turf purposes. More research is needed to overcome these seeding, establishment, and disease hazards. In most grasses, progress in developing improved varieties depends on the accumulation of basic information in the fields of cytology, genetics, pathology, and physiology. Problems associated with management and maintenance of turfgrass areas are poorly understood. More than three billion dollars are spent annually in establishing and maintaining turf. Improved management practices and improved grass varieties are urgently needed to meet the different requirements of use, climate, and soil that characterize the major turfgrass regions of the United States.

USDA AND COOPERATIVE PROGRAM

The Department has a continuing long-term program involving agronomists, geneticists and plant pathologists engaged in both basic and applied research. Forage grass and turf research is under way at Tucson, Ariz.; Tifton, Ga.; Lexington, Ky.; Beltsville, Md.; State College and Stoneville, Miss.; Lincoln, Nebr.; Mandan, N. Dak.; Woodward, Okla.; University Park, Pa.; College Station, Tex.; Logan, Utah; and Madison, Wis. All work is conducted in cooperation with the respective State Agricultural Experiment Station. New work was initiated at Tifton, Georgia, to develop insect-resistant bermudagrass varieties in cooperation with the Entomology Research Division. Research on relationship of plant pathogens to toxicity in tall fescue is being conducted under contract at Lexington, Kentucky.

Four PL 480 projects are active in India; namely, Cataloguing and classifying genetic stocks of Pennisetum spp.; The establishment of linkage groups in Pennisetum typhoides; Pollen storage and serological classification; and Genetic evaluation of grain and fodder quality in millet. In addition, a project has been negotiated to study cytogenetics of grass species in the tribe Andropogoneae. A PL 480 project in Rio Grande do Sul, Brazil, involves ecological and cytological studies and genetic improvement of forage grasses and legumes.

The Federal scientific effort devoted to research in this area totals 16.5 professional man-years. Of this number, 12.5 are devoted to breeding and

genetics; 1.2 to diseases; .9 to quality and variety evaluation; and 1.9 to culture and physiology.

PROGRAM OF STATE EXPERIMENT STATIONS

A total of 129.9 professional man-years is devoted to this area of research.

PROGRESS -- USDA AND COOPERATIVE PROGRAMS

A. Breeding and Genetics

1. A new allotetraploid hybrid combining Old World and New World wheatgrass species. Allotetraploid species ordinarily originate by chromosome doubling of sterile hybrids between two diploid species. Chromosome doubling of two diploid species followed by their hybridization could also give rise to an allotetraploid species, although this mode of origin has not been previously documented. The latter means of allotetraploid synthesis was demonstrated at Logan, Utah, by controlled hybridization of two basically autopolyploid species, Agropyron desertorum and A. spicatum, both $2n=28$. Four hybrids were obtained from this cross. Each hybrid behaved cytologically as an allotetraploid. The F_1 hybrids were sufficiently fertile to permit intercrossing.

2. Sterility overcome in hybrids between quackgrass and crested wheatgrass. Sterility of F_1 hybrids between quackgrass (Agropyron repens) and crested wheatgrass (A. desertorum) has been overcome by chromosome doubling. Meiosis in the hybrids is quite regular, and more than 80% of the pollen is stainable. The 70-chromosome hybrids are morphologically similar to their 35-chromosome counterparts in that they are vigorous, leafy, highly productive, and mildly rhizomatous. Fertile, cytologically stable derivatives of quackgrass X crested wheatgrass hybrids are being evaluated for agronomic potential at Logan, Utah.

3. Genetic basis established for mode of reproduction in buffelgrass. Research at College Station, Texas, demonstrated that apomixis in buffelgrass is controlled by one dominant and one recessive gene pair. Mutation at the recessive locus gives rise to a dominant allele with epistasis which results in a sexual plant. Double recessive is assumed to result in sexual plants. A variant plant discovered in a field of apomictic blue buffelgrass proved to be heterozygous and sexual. Segregation among progenies from F_1 hybrids between this plant and an apomictic one gave conclusive evidence for the validity of the proposed genetic model. Superior obligate apomictic F_1 hybrids have been isolated from hybridization between sexual and apomictic parents. One of these lines which combines desirable characteristics of present commercial varieties but with superior seed and forage characteristics is expected to be released shortly. Because desirable characteristics can be combined and fixed in one generation, the apomictic breeding system now offers potential for production of superior varieties with hybrid vigor.

4. Sonora black gramagrass released. Sonora black gramagrass was released by the Arizona Agricultural Experiment Station in cooperation with the Plant Science Division, Soil Conservation Service, and the Crops Research Division, Agricultural Research Service. Nutritious and drought-tolerant black gramagrass, a major range grass species in Arizona, New Mexico, Southwestern Texas, and adjacent areas of Mexico, is also an important forage grass in parts of Oklahoma, Colorado, Utah, Nevada, and California. Poor and unreliable seed production has limited the widespread use of this long-lived perennial species for range seeding. Sonora is the first improved high seed-yielding black gramagrass to be released for commercial seed production.

5. Kenwell tall fescue released. Kenwell tall fescue, developed at Lexington, Ky., was released in cooperation with the Kentucky Agricultural Experiment Station. The variety is characterized by improved palatability to livestock, improved leaf disease resistance, and better maintenance of color during summer drought as compared to Ky. 31 tall fescue. Because Kenwell is 5 to 7 days later in date of flowering than Ky. 31 and is somewhat less competitive, Kenwell offers a better opportunity for maintaining associated legumes in stands. Comparisons with commercial varieties for palatability, showed that Kenwell was significantly better grazed in the summer and fall by cattle having free choice. No case of fescue-foot toxicity was recorded for Kenwell prior to its release, but livestock developed fescue foot in the summer of 1965 on Kenwell and Ky. 31 pastures at Middleburg, Va.

6. A simplified regression method for estimating rapidity of grass seed germination. A regression index that expresses directly the number of days to achieve 50 per cent germination in unequal size seed samples was developed at Tucson, Arizona. The regression index method is being used in the blue panicgrass breeding program at Tucson, Ariz., with the objective of selecting for rapid and for slow seed germination. The effect of seed germination rates of established populations on seedling drought tolerance is part of a breeding program at Tucson with the objective of ascertaining factors associated with drought-tolerant range grasses.

7. Intraclonal variation in smooth brome and timothy. Intraclonal variation in morphological and reproductive characteristics has been established at Madison, Wisconsin, for polyploid smooth brome and timothy. Panicles are produced at two different plant heights. Timothy seedlings derived from different inter- and intralevel panicles varied in numbers, texture, and vigor. Differences in self-fertility occurred in single panicles of smooth brome obtained from inter- and intraheight categories. Cytological studies with this material are in progress.

8. A new male-sterile pearl millet released. The cytoplasmic male-sterile Tift 18A and its maintainer Tift 18B were released in cooperation with the Georgia Coastal Plain Experiment Station, Tifton, Georgia. Tift 18A was developed by repeatedly backcrossing a previously released male-sterile Tift 23A with Tift 18. The Tift 18 inbred produces fewer stems, usually

three to five, but these are twice as thick in diameter as those produced on plants of Tift 23. In contrast to the blue-gray seeds of Tift 23, Tift 18 produces white seeds borne in heads 18 to 36 inches long. Both inbreds flower in about 90 days and produce seed 3 to 4 weeks later when planted in May at Tifton, Georgia. At maturity, the plants are 6 to 8 feet tall. Because Tift 18A and B carry a dormancy factor that causes poor germination of freshly harvested seed, seed damage is reduced in heads during rainy periods. Tift 18 was released because of the white seed and large heads sought by people in Africa and India who grow more than 35 million acres of pearl millet for its grain. When used in F_1 hybrids, Tift 18 should greatly increase yield and improve quality of the grain products.

9. Additional interspecies Paspalum hybrids obtained. Research at State College, Miss., has shown that interspecific hybrids among Paspalum spp. may be obtained by use of a heavily shaded and high-humidity greenhouse for the emasculation and pollination operations. Low crossability and cytological data from hybrid meiosis indicated little relationship among P. dilatatum, P. pubiflorum, P. platyphyllum, P. virgatum, P. malacophyllum, and P. urvillei. P. dilatatum appeared to be closest to P. urvillei. When obligate apomict P. dilatatum was the female parent, all hybrids developed from fertilization of unreduced eggs and were obligate apomicts. The reproductive behavior of hybrids from reciprocal crosses varied with that of the female parent. The apparent effect of different parental genotypes on expression of mode of reproduction is determined to a great degree by their functional gametes. Cytogenetic analyses at College Station, Texas, suggested homology of one genome for P. dilatatum x P. pubiflorum and P. dilatatum x P. platyphyllum hybrids. The P. urvillei chromosomes were apparently nonhomologous to the P. malacophyllum chromosomes.

10. Increase in soluble carbohydrates by breeding of sorgho x johnsongrass. Progress is being made at Stoneville, Miss., toward selecting for higher total water-soluble carbohydrates among progenies of Hodo sorgho x johnsongrass hybrids. Two lines contain water-soluble carbohydrates that are within 2 per cent of that contained in forage of Sart sorgho harvested at the same growth stage. This high amount of total soluble carbohydrate appears when plants are 4 feet tall and remains nearly constant until maturity. Progress is being made toward combining selections for high carbohydrate content and selections with high dry matter production.

11. Accessory or B-chromosomes found in Cynodon species. Chromosome counts from a number of Cynodon dactylon clones showed 23 clones with $2n=36$, 5 with $2n=36 + 1B$, 9 with $2n=27$, 5 with $2n=18$, and one with $2n=18 + 1$ large accessory chromosome in some cells. Types of C. barberi were found with 18, 27, or 36 chromosomes. Clones of C. coursii and C. polevarisii had 36 chromosomes but no accessory chromosomes. The effect of accessory chromosomes on fertility and plant development is being studied at Tifton, Georgia.

12. Differential in functional gametes from wide crosses in Bromus. The plant types of progenies obtained at Madison, Wisconsin, by selfing partially

fertile hybrid derivatives of octoploid Bromus inermis X B. titholepis were highly correlated with those of their maternal parents. The direction of the original hybridization influenced the plant types recovered in the inbred progenies. A similar influence of the maternal parent was expressed in seed set. Restitution and substitution backcrosses to either the original parents resulted in the recovery of the recurrent parental types. These were recovered in proportions that exceeded those theoretically expected on the basis of random recombination. Similar results were obtained from crosses involving B. pumpellianus. These and similar data point to a hypothesis of biochemical systems operating at the genome level. The chromosome complement of polyploid grasses may be visualized as being composed of a series of such systems. This hypothesis offers an explanation for the frequent failures of obtaining gene transfers in wide crosses in grasses.

13. Refinement in Syn 1 seed production of Russian wildrye varieties.

Erratic seed yields were found to be particularly troublesome in a multiple-clone synthetic wildrye variety. Factors which contributed to the variable performance of Syn 1 seed production on parental clones were seed set, seed size, and seed yield. The effects of these factors were reduced by compositing the Syn 1 generation seed on the basis of equal numbers of viable seed. Further research under way at Mandan, North Dakota, attempts to evaluate the effect of equalizing the pollen contribution in the recombination block by the use of comparable numbers of panicles. Analysis of seed size from reciprocal diallel crosses of Russian wildrye demonstrated a paternal effect ranging from 12 to 40% and which occurred consistently with the same parents in two consecutive years.

14. Progress toward restoring fertility of intergeneric ryegrass-fescue hybrids.

Basic studies at University Park, Pa., involved intergeneric chromosome-doubled (perennial ryegrass x induced tetraploid meadow fescue) x tall fescue trispecies hybrids which have been backcrossed to tall fescue. A wide range in pollen stainability and seed fertility was recorded for progenies from open pollination of selected backcross clones. Mean seed yields from open pollination of polycross progenies from one group of backcross clones which were phenotypically intermediate between the parent species and another group of clones which resembled tall fescue were not statistically different. This suggests that rapid improvement could be made in restoring fertility among progenies from backcross parents which were phenotypically intermediate between ryegrass and tall fescue. The chromosome number for the backcross generation and unselected progenies was stable at the hexaploid level which is a prerequisite for achieving high fertility in this breeding material. Cooperative basic research conducted at Lexington, Ky., has shown that colchicine treatment of sterile 28-chromosome hybrids between annual ryegrass and tall fescue produced partially fertile 56-chromosome amphiploids. Three generations of selection resulted in a high proportion of progenies with 56 chromosomes, but also a few 42- and 49-chromosome derivatives. Approximately 84 per cent of progenies showed anther dehiscence in certain lines from amphiploid parents. Plants with good seed set produced progenies which ranged from completely sterile to

highly female fertile. Low seed set in amphiploids was attributed to complementary gene action inherited from the ryegrass and tall fescue parent. High seed-yielding 56-chromosome derivatives have been isolated and are being used in further breeding work at Lexington, Ky. Likewise, fertile 42-chromosome derivatives from the amphiploids have been isolated to study effect of different levels of ploidy on the stabilization of agronomically desirable populations derived from intergeneric hybrids.

15. Pearl millet varieties and inbreds evaluated in India. PL 480 research in Ludhiana, India, employed multiple correlations to compare efficiency of selection indices in breeding of pearl millet using 30 Indian varieties, 70 Indian inbreds, 25 African varieties, and 50 American inbreds. Selection indices based on leaf size, leaf number, tiller number, plant height, and earliness accounted for 44 to 79 per cent of variation in green weight forage yield. Selection indices based on grain size, grain hardness, plant height, tillering, and earliness accounted for 38 to 82 per cent of total variation in grain yield. The genetic variability for protein content ranged from a low of 45.1 per cent of total germ plasm for American inbreds, to a high of 66.7 per cent for Indian inbreds. Correlations of protein content with grain characters studied were nonsignificant. Further work under this project includes chemical analyses and determination of yield and quality of forage from hybrids between napiergrass and selected pearl millet lines.

16. Nonflowering orchardgrass for high quality forage. Orchardgrass varieties adapted to the Northeastern United States generally flower when exposed to a period of low temperatures. Plants selected at University Park, Pa., remained nonflowering throughout the growing season or produced very few flowering stems. In Western States, these same plants flowered and produced seed. Seed produced at Prosser, Wash., from one 4-clone synthetic averaged 1.2 panicles per square foot before first cutting as compared to 25 panicles for Pennlate in field tests in Vermont and Pennsylvania. The low panicle production resulted in reduced forage yield; but the synthetic had less lignin, cellulose, and hemicellulose as compared to Pennlate. This nonflowering synthetic shows promise in producing high quality forage by retaining, over a long growth period, a low ratio of stems to leaves.

B. Diseases.

1. Fungicide treatment controls fairy ring. Captan 50-W rototilled into soil at 5 pounds per acre was the only pesticide of 5 tested that controlled turf fairy rings for 3 years in plots of centipede grass at Tifton, Ga. Fore, Manzate-D, Dithane Z-78, and Panogen inhibited fairy rings for 3 months when applied with surfactants. With the exception of Captan 50-W rototilled into the soil, available pesticides appear unsuitable for the homeowner.

2. Management practice has little effect on alkaloid production in tall fescue. In order to better understand the nature of fescue toxicity to cattle, contract research was conducted at Lexington, Ky., on the accumulation

and types of alkaloids in tall fescue. Total plant and ergot alkaloids were measured by thin layer chromatography. Different plant management practices had little or no effect on the occurrence of plant alkaloids. The number and quality of alkaloids varied with season and increased during the summer. Three unidentified plant alkaloids were discovered in tall fescue, but one of these was not observed in annual ryegrass x tall fescue hybrids. The accumulation of ergot alkaloids indicated an orderly sequential synthesis in sclerotia, starting with ergometrine and followed by ergotamine, ergo-cornine and/or ergocristine. Accumulation of ergot alkaloids had no apparent effect on accumulation of plant alkaloids in ergot-infected tall fescue. Information is still needed on the role of alkaloids in fescue toxicity.

C. Culture and Physiology.

1. Good quality Kentucky bluegrass turf maintained by an annual application of nitrogen. At Beltsville, Md., plots of Kentucky bluegrass turf which received nitrogen fertilizer both in the spring and fall had significantly more leafspot disease than those receiving either spring or fall only applications. Although there was a definite trend for less leafspot with fall applications of nitrogen, the results were not significantly different from spring application. More disease resulted when organic nitrogen sources at 2- and 4-pound rates per 1,000 square feet were applied in the fall rather than in the spring. In contrast, plots fertilized with 1 and 2 pounds of ammonium sulfate in the spring had more disease than plots fertilized in the fall. Satisfactory turf density was obtained with 2 pounds of nitrogen applied either in the fall or spring.

2. Bentgrass turf varieties differ in reaction to herbicides for Poa annua control. At Beltsville, Md., injury to bentgrasses from preemergence application of herbicides was generally specific to the variety. Washington bentgrass was severely injured by a 12-pound per acre application of Siduron. Calcium arsenate caused injury to Old Orchard. Slight injury was observed on Cohansey bentgrass when treated with DCPA at 10 pounds per acre. In greenhouse tests, complete control of annual bluegrass at levels of phosphorus ranging from 0 to 3492 pounds per acre and at two planting dates was obtained with trifluralin at 2 pounds per acre. For both planting dates, high levels of phosphorus reduced the effectiveness of both calcium arsenate and, to a lesser degree, DCPA. Although DMPA was more effective than bensulide for the control of annual bluegrass on a plant count basis, bensulide was more effective on a yield basis.

3. Irrigation and fertilization improve seed production from established stands of sideoats grama. Irrigation in the fall and at heading time plus 40 pounds of nitrogen per acre resulted in greatest number of racemes per linear foot of sideoats grama grown in 40-inch rows in Nebraska. The 40 pound rate which averaged 2.25 pounds of clean seed per pound of N applied yielded more on a pound-of-N-basis than did the 80 and 120 pound rates. Seed quality improved with fall and heading irrigation but was poorest with fall only irrigation. Both 40 and 80 pounds of N resulted in significant

improvement in seed quality. Water use efficiency was lowest when only one irrigation was applied in the fall and highest when water was applied in the fall and again at heading. Nitrogen fertilizer significantly increased water use efficiency with the first 40-pound increment having the greatest effect.

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PASTURE AND RANGE QUALITY, PHYSIOLOGY, SEEDING
AND ESTABLISHMENT AND MANAGEMENT
Crops Research Division, ARS

Problem: Grazing lands of the United States occupy almost one billion acres as compared to 350 million acres of all harvested crops. Presently more than half of all the nutrients consumed by domestic livestock come from pastures and ranges. With increasing population and increased demand for livestock products, the proportion of nutrients from pastures and ranges is expected to increase. Grazing lands represent a highly diverse part of our land resource and, because of their diversity, are a tremendous challenge to research. More precise information is required for many situations; information about what plants or mixtures meet the requirements for yield, nutritional value, and resistance to heat, cold, drought, and pests. Some of the major categories needing research attention are concerned with seeding and establishment, and include basic and applied physiological and ecological studies on the response of pasture and range species, biochemical constituents, and morphological development of the plant under grazing. In future years, as humans compete more intensively with livestock for present feed grains, we must develop our grassland resources if this country expects to maintain high quality diets with the levels of meat and milk that are considered essential.

USDA AND COOPERATIVE PROGRAM

The Department has a continuing long-term program involving agronomists, plant physiologists, range scientists, and chemists engaged in basic and applied research on the management and improvement of grazing lands, native meadows, and improved pastures. All work is cooperative with the respective States, with the U. S. Forest Service, and Bureau of Land Management, in areas where grazing is integrated with lands under their control. Research is in progress at Flagstaff, Ariz.; Berkeley, Calif.; Ft. Collins, Colo.; Gainesville, Fla.; Tifton, Ga.; Twin Falls, Idaho; Lafayette, Ind.; Beltsville, Md.; St. Paul, Minn.; Columbia, Mo.; Bozeman, Mont.; Miles City, Mont.; Reno, Nev.; Las Cruces, N. Mex.; Ithaca, N. Y.; Raleigh, N. C.; Mandan, N. Dak.; Woodward, Okla.; Burns, Oreg.; University Park, Pa.; Logan, Utah; and Pullman, Wash. Research at Bozeman, Mont., and Mandan, N. Dak., is conducted cooperatively with Soil and Water Conservation Research Division and at Miles City, Mont., with Animal Husbandry Research Division.

Of four P.L. 480 projects, two are in Israel. One concerns developmental physiology of perennial pasture grasses. The other investigates establishment and maintenance of seeded dryland range under semiarid conditions. A project in India concerns basic studies on polyfructosan biosynthesis in fodder crops. A project in Poland concerns evaluation of the ornithine cycle in higher plants.

The Federal scientific effort devoted to research in this area totals 32.2 professional man-years. Of this number, 5.3 is devoted to quality; 10.2 to physiology; 6.3 to seeding and establishment; 0.3 to processing; and 10.1 to management.

PROGRAMS OF STATE EXPERIMENT STATIONS

A total of 60.0 professional man years is devoted to this area of research.

PROGRESS -- USDA AND COOPERATIVE PROGRAMS

A. Quality

1. Laboratory technique predicts in vivo digestibility and voluntary intake. Digestibility of 12 forages was determined at Lafayette, Indiana, using the in vitro technique. Incubation for 48 hours, with rumen fluid plus pepsin, was highly correlated with in vivo dry matter digestibility ($r = .97$). An incubation period of 6 hours with rumen fluid plus pepsin was highly correlated with voluntary intake ($r = 0.87$). In vitro digestion with rumen fluid only resulted in separate prediction equations for the grasses and legumes. The additional pepsin digestion apparently resulted in greater solubilization of the legume dry matter and permitted a single prediction equation for both grasses and legumes.

2. The meadow vole, a possible bioassay for forage quality. The meadow vole is a small herbivore which can utilize fibrous herbage. Samples of ground alfalfa, fescue, and brome grass hay were fed to weanling voles at Lafayette, Indiana. Intake and digestibility results were compared with in vivo feeding trials with sheep. The relationship with dry matter intake was highly significant ($r = .75$). However, the relationship with digestible dry matter was low ($r = 0.32$). Further work with nonsupplemented forage diets is planned to determine the usefulness of the meadow vole to bioassay for forage quality.

3. Kentucky 31 tall fescue out performs Kenwell in grazing trials. At Beltsville, Maryland, 4 years of grazing trials with beef cattle show that daily gains were 0.9 and 0.7 pounds for Kentucky 31 and Kenwell, respectively. Kentucky 31 produced 100 pounds more beef per acre per year than Kenwell (314 lbs. vs. 211). Cattle consumed slightly more of the more palatable Kenwell forage, but the Kentucky 31 carried slightly more animals per acre.

4. Forage quality in bermudagrass is inherited. At Tifton, Georgia, the nylon bag dry-matter-digestibility technique suggests that strains of bermudagrass differ in digestibility. Coastal x Kenya No. 14 had an average digestibility of 58% as compared with 52% for Coastal. If one assumed equal consumption of dry matter by an 800 pound steer, daily gains on the Coastal x Kenya hybrid would be 1.47 pounds per day or 30% greater than that for Coastal bermudagrass.

5. Improvement in technique for lignin determination. At least two substances, a lipid and a protein, contribute to the apparent lignin content of forage. As a result of studies at University Park, Pennsylvania, changes in the technique for determining lignin were made. Addition of a wetting agent, hexadecyltrimethylammonium bromide, had no affect on the activity of pepsin during digestion and made unnecessary higher than room temperature digestion. The wetting agent increased the amount of protein removed and contributed to more reproducible results. The practice of washing with concentrated hydrochloric acid to remove some of the carbohydrate material was found to be nonessential and was eliminated. The changes increased the speed and reproducibility of the analysis for lignin.

6. New approach to the determination of forage quality under grazing. Forage quality can be determined from data obtained by extension of a new method of determining feed intake, according to research at Ft. Collins, Colorado. In addition to measuring water drunk and water in the forage consumed, the method requires determination of distance traveled and animal weights at start and end of test. An adaptation of the net energy equation for maintenance and production, combined with the above data, provides a reliable, simple estimate of forage quality.

7. Russian wildrye superior to native range for fall grazing. In studies at Mandan, North Dakota, digestibility of herbage from pastures of Russian wildrye and native range (deferred for late fall grazing) was determined on sheep-fed pelleted rations. Russian wildrye dry matter was 46% and the native range 34% digestible. Percentage protein, calcium and phosphorus of Russian wildrye herbage was higher than that of comparable native grasses; and the fiber and lignin were lower. Steers were grazed on both kinds of pasture for an average of 52 days for each of 3 years. Average gain per head on the Russian wildrye was 23 pounds, but steers on native grass lost 5 pounds per head during the same period. There were high positive correlations between digestibility, intake, and gains. Russian wildrye is clearly superior to native range for late fall and early winter grazing.

B. Physiology

1. Environment regulates RNA metabolism, protein synthesis. At Gainesville, Florida, growth regulator-induced metabolic changes have been compared with those changes which are observed when growth is adversely affected by environmental factors. Ribosomes, upon which protein is polymerized, are not seriously affected by drought or temperature extremes, but the synthesis of messenger RNA (mRNA) may be the factor which is modified by environment and controls growth.

2. Photoperiod controls stem formation on roots. At Beltsville, Maryland, plants of creeping-rooted alfalfa, sheep sorrel, and crownvetch showed similar morphologic and physiologic responses when grown at photoperiods of 8, 11, 14, and 17 hours. Generally, the critical daylength for the long-day response was between 11 and 14 hours. Plants exhibiting the long-day

response, produced more nodes, longer internodes, greater top weight, but fewer adventitious stems on roots. The long-day inhibition of stem production on roots was inversely associated with top growth and appears to be dependent upon it. It is suggested that daylength influences stem initiation on roots through the interrelationships of auxins produced in the tops and kinetin-like substances produced in the roots.

3. Ten morphologic stages define growth in white clover. Daily ratings of morphologic stages of development of ladino clover leaves are a simple and rapid technique for following leaf growth. This system of rating development of leaves has been used at Beltsville, Maryland, to follow rate of leaf development as affected by environment, management, or as an identifying point for initiating physiologic treatments on clover seedlings.

4. Foam test reliable index of bloat potential in legumes. In studies at Bozeman, Montana, foam production of 27 native and introduced legumes was proportional to their known bloat potential. Low-foaming legumes included Lotus corniculatus and Onobrychis viciaefolia, known to be essentially non-bloating; Trifolium pratense, T. repens, Medicago sativa, and Melilotus sp., known to be bloat hazards, produced 20 to 50 times the volume of foam.

5. Polyfructosan biosynthesis in fodder crops. This P. L. 480 project in India was terminated without reaching the main objective. However, results of the study will be useful in providing background information, methods for further research, and information on the presence or absence of various carbohydrates in several plants.

6. The ornithine cycle in higher plants. Work on this P. L. 480 project in Poland is directed towards relating metabolism of the ornithine cycle to biosynthesis of pyrimidine nucleotides. Observations to date show that carbamyl phosphate rather than CO₂ is the near precursor in pyrimidine biosynthesis.

C. Seeding and Establishment

1. Subterranean termites threat to range seedings in Southwest. Recent studies at Logan, Utah, have shown that in areas where winter temperatures rarely drop below 10°F and average January temperature is above 30°F subterranean termites may be a major factor in the destruction of natural seed supplies on rangelands.

2. Cheatgrass aggressive reseeder on western rangelands. Cheatgrass (Bromus tectorum) provides tremendous competition to seeded perennial grasses, particularly in the seedling stage. In a fall favorable for cheatgrass germination, more than 400 plants may emerge per square foot of land. Research at Logan, Utah, has shown that under such conditions the emerged seedlings represent less than 50% of the total viable seed population.

3. Low temperatures retard germination of some forage grasses. Research in

Israel on a P. L. 480 project showed that perennial forage grasses of value in range seeding in the Western United States are sensitive to and retarded in germination by low temperatures in contrast with cereal grains and some annual forage legumes.

4. Old plantings verify adaptation of crested wheatgrass. Plantings in southern Idaho ranging from 20 to 32 years old in the sagebrush type have clearly demonstrated the adaptation of crested wheatgrass (principally Agropyron desertorum) to arid lands of the Intermountain region, according to research headquartered at Logan, Utah, and Twin Falls, Idaho. With reasonable management, crested wheatgrass has not died out as a result of drought, heat, cold, fire, or disease. It is more resistant than native species, to fire and heavy grazing, and is more aggressive in natural spread from seed.

D. Management

1. Row direction influences blue lupine yield. At Gainesville, Florida, four years of testing shows that east-west rows of blue lupine outyield north-south rows by about 13%. Better light interception, soil moisture, and temperature have contributed to the increase in yield. Soil temperatures at midday were as much as 10° cooler and relative humidity 8% higher in east-west rows than in north-south rows.

2. Grazing management differs for reed canarygrass and brome grass. When pastures in Minnesota were grazed lightly, reed canarygrass produced 100 pounds more beef per acre than brome grass. However, under heavy grazing pressure, brome grass out-produced reed canarygrass by 80 pounds of beef per acre.

3. Concentrate feeding to low-producing dairy cows uneconomical on excellent pasture. In Minnesota, dairy cows receiving 11 pounds of concentrate per day on brome grass pastures produced only 3 pounds of milk per day more than cows not receiving concentrate (27 vs. 24 pounds). This represented a net loss of 13 cents a day for concentrate feeding.

4. Tree spacing affects beef production in slash pine pastures. At Tifton, Georgia, tree spacing significantly affected pounds of beef per acre; live-weight gains of 305, 236, and 132 were obtained from pastures containing no trees and trees spaced on 20 and 12 foot centers, respectively. Average daily gains did not differ for Coastal bermudagrass, prostrate dallisgrass, Pensacola bahiagrass, and carpetgrass.

5. Grazing management of mountain pastures. Average daily gains were lowest on nitrated mountain pastures in North Carolina, but increased carrying capacity resulted in greatest amount of beef per acre. Grazing of alfalfa and birdsfoot trefoil pastures in addition to mountain pastures resulted in daily gains and beef production per acre which was higher than on mountain pastures alone. Mountain pastures over-seeded with birdsfoot trefoil

generally produced high average daily gains but low carrying capacity. It seems reasonable to combine nitrated mountain pastures with birdsfoot trefoil or alfalfa to increase midsummer carrying capacity and overall forage and beef production.

6. Forage from lead-contaminated soil not toxic to animals. Bromegrass plants grown on soils contaminated with lead (680 p.p.m.) from a nearby battery smelter in Minnesota did not accumulate sufficient quantities of lead to be toxic to animals. It is reported that at least 150 p.p.m. lead in the total ration is needed to be toxic, and the greatest concentration accumulated in the forage was only 12 p.p.m.

7. New approach to measuring feed intake under grazing. The relationship of water intake to feed consumed has been correlated to determine the dry matter consumed by grazing animals at the Central Plains Experimental Range at Nunn, Colorado. Other essential data concerns air temperature and percent moisture in forage grazed. The method is promising at temperatures below 90°F and with forage that fails to provide the grazing animals' water needs.

8. Spring burning beneficial to shinnery oak rangeland. Research at Woodward, Oklahoma, over the past 5 years has shown that controlled burning of shinnery oak ranges not only increased yield but improved both the palatability and availability of the forage. In dry years, burning in April gave greatest benefits, while in wet years burning can be extended to mid-May. Increase in forage yield, in response to controlled spring burning, has ranged up to 500 lbs/acre.

9. Fertilizer profitable on seeded grasses in Southern Plains. At Woodward, Oklahoma, application of 35 lbs. N/acre each May to pastures of weeping lovegrass and Caucasian bluestem moderately grazed yearlong, increased forage production 40%, carrying capacity 25%, gain per steer 10%, and beef per acre 40%. Actual beef production per acre was 205 pounds on fertilized grass vs. 145 pounds on nonfertilized (3-year average). Summer rainfall during the 3 years was about 80% of normal. Fertilization increased net profits approximately \$2.50/acre/year.

10. Time element important when weighing experimental cattle. Studies at Woodward, Oklahoma, have shown significant weight changes in relatively short periods when cattle are held off feed during weigh-days. Twenty steers lost an average of 6 lbs/head 85 minutes after the first weighing and 10 lbs/head 3 hours after the first weighing.

11. Good management practices additive on Southern Plains. Four single practices that increased beef production in studies at Woodward, Oklahoma, totalled 92 lbs. gain/steer, while the four all applied simultaneously led to steer gains of 87 lbs/head. Proper stocking rate (7 acres/steer, compared with 5 acres) increased gains 9 lbs/steer; 3 lbs. cake/head/in winter, compared with 1.5 lbs/head increased gains 21 lbs/steer; 1 lb. cake/head/daily in late summer vs. none yielded 16 lbs. gain; and steers treated

with stilbestrol gained 46 lbs. over those not treated.

12. Low early season production supports spring deferment of native range. At Mandan, North Dakota, only 1.5 and 8.6% of the season total dry matter on native rangeland was produced by May 1 and May 15, respectively. This was determined by sampling at 2-week intervals during each growing season for 8 years. The August 1 yield was used as 100% production. The June 1 and June 15 percentages were 20 and 39, respectively. Over 70% of the total dry matter for the season was produced by July 1. The yield of new growth in pounds dry matter per acre when mowed at 1-inch above the ground averaged 24, 139, 331, 637, and 1194 for the biweekly samples cut May 1 through July 1.

These data emphasize the importance of deferring grazing on northern plains range until at least mid-June. Earlier grazing removes the small amount of new growth in preference to the dry grass left from the previous year, resulting in a reduction of total production for the season.

13. Crested wheatgrass and native range are complementary in production and use. Crested wheatgrass pasture is an excellent supplement to native range in the Northern Plains primarily because of its early season availability. Studies at Mandan, North Dakota, show that production of native range can be increased approximately 50% by grazing crested wheatgrass up to July 1 and deferring use of native range until after July 1. Even though the total dry matter per acre for crested wheatgrass and native range was similar, gains per acre were much higher on crested because of the high intensity of grazing that could be used. Gains per acre were 50% higher on crested wheat during a period of 45 days in the spring than on native range grazed over the entire summer period of 140 days. Gains up to July 1 on crested wheatgrass were 101 pounds per acre compared to only 21 pounds up to this date on native range. Steer days grazing per acre were also 50% higher on the crested wheatgrass. Consumption of dry matter per acre was 88% higher on crested wheatgrass. Pounds of dry matter consumed per pound of gain was 9.93 and 12.60 on crested wheat and native, respectively. The lower requirement on crested wheat was due to gains early in the season when the grass was green and more highly digestible.

14. Root growth responds less than top growth to N on Northern Plains. Nitrogen applications to rangeland at Mandan, North Dakota, consistently increased yields and most of the response was in top growth. Application in the fall of 30 lbs. N/acre led to an increase in yield up to 1227 lbs/acre. The corresponding weight of roots, to a 4-foot depth, was increased only 2.45 grams. Intensity of grazing had no significant effect on weight of roots in the 4-foot zone, but heavy grazing tended to concentrate roots nearer the surface.

15. Prolonged heavy range use has multiple ill effects. At Miles City, Montana, neither the immediate nor the long-time interests of ranchers are served when rangelands are subjected to continued heavy use. This practice

reduced the fertility of beef cows (and hence lowered the calf crop), the rate of calf growth, the forage yield, the carrying capacity, particularly during drought years, the water infiltration rate and hence water storage in the soil, and return per acre. Continued heavy use increased unpalatable plants, soil compaction, run-off, soil erosion, death losses and need for expensive supplemental feeding.

16. Intake more important than digestibility of reed canarygrass. Studies on flood meadows at Burns, Oregon, have shown the digestibility of reed canarygrass to be relatively high, although it declines like all forage plants as the season advances. Poor animal performance is believed to be associated with limited intake and low percent protein. On June 9, (the boot stage) protein content was 10.25%, while a month later it had dropped to 5.44%.

17. Time of mowing important in weeping lovegrass management. At Woodward, Oklahoma, weeping lovegrass mowed in September and October permitted some fall regrowth, resulting in weakened stands that were reduced by winter killing. When mowing was deferred until November, no injury resulted. Stand reduction from early mowing ranged from 10 to 50% and forage yields from 25 to 50%. Some loss of vigor was also noted following mowing during April.

18. Nitrogen changes botanical composition of native range. Research at Mandan, North Dakota, showed that regular fall applications of N to native range lead to an increase in western wheatgrass and a decrease in blue grama. Eight years of N application at 0, 40, 80, and 160 lbs. N/acre changed the number of western wheatgrass stems/4 sq. ft. from 28 to 11, 84, 156, and 189, respectively. The corresponding changes in blue grama density from 36% in 1958 were 40, 34, 24, and 15, respectively. Under the treatment imposed, western wheatgrass increased rapidly, 150% the first season, while the decline in blue grama occurred slowly.

19. Yields of tobosa highly variable on Southwestern floodplains. Yields of tobosa grass may vary from a low of less than 100 pounds forage/acre in drought years and on sites not receiving run-in, to as much as 2591 lbs/acre in favorable seasons accompanied by run-in, according to studies conducted on the Jornada Experimental Range near Las Cruces, New Mexico. Where run-in occurs, further significant increases in yield are obtained from use of nitrogen fertilizer.

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SEED CROP CULTURE, PHYSIOLOGY, DISEASES, AND VARIETY EVALUATION
Crops Research Division, ARS

Problem: Domestic production of many grass and legume seed crops is 10 to 30 percent below national consumption. To overcome deficits and to assure adequate supplies, 75 million pounds of forage-crop seeds are imported annually. This is necessary because technology has not been developed to make seed production a profitable farm enterprise. Research is needed to investigate the factors that limit tiller initiation, transition from the vegetative to the reproductive growth, fertilization and seed formation, and to develop cultural and management techniques including disease control practices that will assure efficient seed production. Fundamental physiological investigations on growth responses of grasses and legumes to various environmental, management and microclimate factors are needed to establish the specific requirements for reproduction. Normal vegetable seed supply is adequate, but geographic concentration of the seed industry is potentially hazardous. Mechanization of the vegetable industry requires increase of speed and uniformity of seedling development, but improvements in processing quality of vegetables often decreases seed quality. There is insufficient basic knowledge of physiology of seed development and germination as these influence the crop-producing potential of seeds and potential industry mechanization.

USDA AND COOPERATIVE PROGRAM

The Department has a continuing long-time program on grasses and legumes involving agronomists, physiologists, and pathologists engaged in both basic and applied research. Investigations are underway at Shafter, Calif.; Lafayette, Ind.; Stillwater, Okla.; Corvallis, Ore.; Logan, Utah; and Prosser and Pullman, Wash. All work is conducted in cooperation with the respective State Agricultural Experiment Stations and the Entomology and Agricultural Engineering Research Divisions.

Basic research on physiology and biochemistry of vegetable seed germination and seedling vigor is carried out at Fort Collins, Colorado, in cooperation with Colorado State University. At Logan, Utah, in cooperation with the State Agricultural Experiment Station, studies involve seed quality as controlled by environmental conditions, including cultural and harvesting practices. Fort Collins and Logan programs are closely coordinated.

Research is being conducted under eight P. L. 480 contracts. In two projects in Finland (not running consecutively), one in Israel, and one in Germany, studies are coordinated with the broad research objective of investigating the influence of environment and management practices on population shifts in cross-pollinated forage-crop varieties when seed is produced in areas outside the regions of origin. In India, research includes

physiology of development of ovules and other reproductive organs and early production of ascorbic acid and ascorbic acid oxidase and the metabolism of bound forms of nicotinic acid (niacinogen) in germination. In Israel, submicroscopic structural changes in mitochondria and other cellular structures are studied in close cooperation with Fort Collins scientists.

The Federal research in this area totals 13.5 professional man-years. Of this number, 5.5 are devoted to Culture; 5.1 to Physiology; 1.0 to Diseases; and 1.9 to Quality and Variety Evaluation.

PROGRAM OF STATE EXPERIMENT STATIONS

A total of 19.1 professional man years is devoted to this area of research.

PROGRESS--USDA AND COOPERATIVE PROGRAMS

A. Culture

1. Alfalfa varieties vary in attractiveness to pollinators. At Logan, Utah, white-flowered alfalfa was equal to purple-flowered alfalfa in attracting nectar-collecting honeybees, but less attractive for pollen-collecting, leaf-cutting bees. Cross-pollination was higher for honeybees than leaf-cutting bees (45.4% vs. 41.2%), but seed production was higher for the leaf-cutting bees (315 grams vs. 265 grams per plant). Cross-pollination but not seed yield was affected by the bee x variety interaction. This highly significant interaction occurred because cross-pollination of the white-flowered alfalfa paired with one of the purple-flowered alfalfas pollinated by leaf-cutting bees was low, but high when pollinated by honeybees. The percentage crossing on spaced white-flowered alfalfa plants as determined by colored hypocotyls was 54.9%, compared with 74.4% for hand-crossed seed without emasculation. Comparisons of white- and colored-flowered alfalfa for percentage of stainable pollen, nectar-sugar per flower, and pollen extrusion were not significantly different.

2. Management practices affect red clover seed yields. At Prosser, Wash., the effect of clipping on seed yield and seed-yield components was determined on two- and three-year-old stands of Altaswede, Dollard, Kenland, and Pennscott varieties. Unclipped plots and plots clipped on May 1 produced higher seed yields than plots clipped on May 21 and June 11. Reduction in seed yields, percent flowering, and number of heads were greater for Altaswede and Dollard than the other two varieties with progressively later dates of clipping. Slower recovery of single-cut types and lower seed yields from the faster recovering early types were suggested as the reasons for the differential effect of clipping on these varieties. Heads in full bloom on July 5, July 19, and August 2 were collected at maturity. The number of florets per head, seeds per head, and seed weight were significantly lower for the later dates of sampling.

3. Cool-season grasses respond to row spacings and fertilizer applications.

The influence of 12-, 24-, and 36-inch row spacings, and 60, 80, and 100 pounds of N per acre on seed production characteristics of five cool-season grasses was studied at Pullman, Wash., for a five-year period. Orchardgrass produced the most seed when planted in 36-inch rows, while Kentucky bluegrass was best in 12- and 24-inch rows. Crested wheatgrass and smooth brome grass seed yields were higher in 24-inch rows, but the 24-inch rows were not significantly better than 36-inch rows. Seed yields of red fescue were not influenced by row spacing. Seed yields of Kentucky bluegrass and orchardgrass were higher when fertilized with 80 and 100 pounds of N per acre than at 60 pounds. Red fescue and smooth brome grass seed yields were not influenced by nitrogen rates, while crested wheatgrass responded only to the higher rate. There was an interaction between row spacing and nitrogen rates with the Kentucky bluegrass and crested wheatgrass only.

4. Influence of depth and duration of burial in soil on longevity and dormancy in grass and legume seeds.

Extended longevity and dormancy of seeds buried in the soil contribute to varietal contamination in seed crops. At Corvallis, Ore., grass and legume seeds buried for 50 months gave these results: (1) Seeds of annual and perennial ryegrass were longer lived in heavy soil with impeded drainage than in lighter, better drained soils. (2) No live seeds remained of orchardgrass, Newport Kentucky bluegrass, Alta tall fescue, chewings fescue, and perennial ryegrass (lighter soil site). (3) Appreciable numbers of viable seeds of annual ryegrass, Highland bentgrass, red clover, and crimson clover were recovered from the soil. In relation to the numbers of seeds buried, annual ryegrass germinated 4.9%, and 0.5% remained dormant; Highland bentgrass germinated 3%, and 2.1% remained dormant; red clover germinated 33.5%, and 14.5% remained dormant; crimson clover germinated 0.35%, and 6% remained dormant. (4) Seeds of clovers and annual ryegrasses, generally, were longest lived when buried at 7-inch depth. The most viability in the Highland bentgrass seeds was found at intermediate burial depths.

5. Aging of stand reduces efficiency of switchgrass seed production.

In a study at Stillwater, Okla., various management practices were used in an established field of Caddo switchgrass to rejuvenate seed production. Treatments included fall and spring burning of crop residue, residue mulching, chiseling 18 to 22 inches deep between the rows, and normal cultivation. Nitrogen was applied each year and irrigation water was supplied to avoid plant stress. Seed yields, however, continued to decline throughout the six years of the study. In 1960, the seed yield was 582 pounds per acre; in 1965, 95 pounds per acre. Data indicate that soil and climatic conditions of a particular year may exert a greater influence than age of stand on yield. A high percentage of the seed harvested in the 4th, 5th, and 6th years was blasted. In 1965, seed loss due to blasting was estimated from 208 to 262 pounds per acre. Seed blasting in switchgrass is generally uniform within a given year and, therefore, may not be directly related to age of stand.

6. Distribution of foundation seed of forage species. Distribution of seed of superior forage-crop varieties by the Foundation Seed Project amounted to 322,337 pounds in 1965. This was an increase from the previous year. Foundation seed was distributed to 12 States and Canada. A total of 460,676 pounds of foundation seed was produced in California, Idaho, Montana, Nevada, New York, Oklahoma, Oregon, and Washington. The foundation seed supply of each variety is adequate to meet expected demands in 1966 and to provide reasonable carry-over reserves.

B. Physiology

1. Breaking seed dormancy increases reliability of germination tests. Techniques have been developed at Stillwater, Oklahoma, to break seed dormancy in Panicum virgatum and P. anceps. Instead of chilling seed for several weeks, dormancy in Kanlow switchgrass, Panicum virgatum, can be broken by soaking seed for 30 minutes in sodium hypochlorite prior to germination. The sodium hypochlorite destroys the outer chartaceous indurate lemma and palea surrounding the caryopses, and possibly scarifies the inner seedcoat. Results are similar to those obtained earlier for Panicum anceps. Whereas 30 minutes were required for P. virgatum, 6 hours soaking were required for seeds of P. anceps. Six to ten weeks pre-chill is required to stimulate germination of P. anceps seeds. Seed respiration studies indicate that oxygen uptake increases rapidly between the fourth and fifth week of pre-chill. Twice as much decarboxylase was active in seeds pre-chilled for six weeks (.056 m/moles/hr/mg protein) as compared to untreated seed (.035 m/moles/hr/mg protein).

2. Short-lived characteristic of red clover may be due to physiologic breakdown. Histological studies at Prosser, Washington, have shown that groups of cells in the pitch of the crown deteriorate when red clover suffers "internal breakdown." Some of the cells are enlarged, others collapse. Starch grains are often found in collapsed cells, so starvation does not appear to be a primary factor. The deteriorated cells are characterized by yellow-brown discoloration and a strong affinity for basic dyes. Neighboring cells apparently remain alive as revealed by tests with tetrazolium salts. In young plants, cells surrounding areas of breakdown often display meristematic activity. The groups of deteriorated cells scattered through the large, highly vacuolated cells of the pith give the appearance of a general "rot." Several stain combinations have been effective in differentiating the affected tissue. Azure B is of special value because of its strong specificity for the affected cells.

3. Environmental requirements for the development of floral stems in red clover varies with ecotypes. At Lafayette, Indiana, over 90 percent of the plants of Pennscott, a double-cut, medium-early variety, developed floral stems within five weeks after seeding when grown under a 16-hour photoperiod and 30° C. Lengthening the photoperiod to 20 hours reduced the time to four weeks. Flowering of 90 percent of the plants of Tammisto, a single-cut, late variety, did not occur until after 10 weeks when grown

under a 16-hour photoperiod and 30° C. A small percentage of the Tammisto plants began developing floral stems after five weeks under a 16-hour photoperiod and four weeks under a 20-hour photoperiod. Lengthening the photoperiod from 16 to 20 hours did not materially affect the percentage of plants that developed floral stems unless plants were exposed previously to low temperatures. Most rapid floral development occurred in Tammisto after the plants were exposed to 2° C. for two weeks, either immediately following seeding or after a two-week exposure to a 12-hour photoperiod followed by a 20-hour photoperiod at 30° C. The 2° C. exposure was slightly more effective than 10° C. From a seed production standpoint, it is significant that under the regimes which included low temperature exposure, plants of most varieties flowered within two to three weeks of each other. This compares with a 5- to 8-week period when plants received no low temperature exposure.

4. Submicroscopic cellular structural changes in germination. At Jerusalem, Israel, under P. L. 480, it has been found that if embryonic axes are allowed to imbibe water for up to 6 hours and are then redried, the fine structure of the cells will survive drying and the axes will retain viability. Beyond this time, redrying destroys both cell structure and viability. These observations suggest that cells of seeds contain some mechanism to protect cellular structure, thus permitting cells to survive desiccation.

C. Diseases

1. Seed disinfection by aerated steam continues to show promise. At Corvallis, Oregon, aerated steam eradicated diseases in seed of several species of grasses and legumes. Agrostis canina seed were least tolerant, and sudangrass seeds were the most tolerant of several species of grasses studied. Pathogens affecting grass seeds also vary in tolerance to steam treatment. Spores of Ustilago avenae and U. bullata in indurate spore masses were killed in 20 minutes at 140° F. Gloeotinia temulenta, the pathogen causing blind-seed disease of ryegrass, was killed in pseudo-sclerotia by 20 minutes exposure at 140° F. Sclerotinia borealis in sclerotia and grass seed nematode in bentgrass seed galls were also eradicated by the same treatment. Aerated steam appears promising for decontaminating grass seed stocks where some loss in seed germination is acceptable. The most favorable temperature for treating grass seed appears to be 140° F. Astragalus cicer was the most tolerant legume species to aerated steam of all species tested. The general high level of tolerance of forage species to aerated steam before loss of germination occurs will permit eradication of nearly all fungi from seeds. Alternaria porri was eliminated from infected carrot seeds with only 10 minutes at 140° F., a treatment that increased the seed germination percentage slightly.

2. Species of smut and other diseases attack grass-seed crops. Stripe smut, Ustilago striiformis, was noted for the first time in a Merion Kentucky bluegrass seed field at Corvallis, Oregon. Although the incidence was only a trace, the disease is considered a potential threat to grass seed production because of adult plant infection. Flag smut, Urocystis agropyri, was identified for the first time also in Merion Kentucky bluegrass. Since the disease occurred in a field which produced foundation seed for several years, it is highly possible that the smut was disseminated to other newly established seed fields. Bacterial blight was discovered in a field of Akaroa orchardgrass. Dwarf bunt occurred in a foundation Potomac orchardgrass seed field grown in Union County. New diseases require a continuing surveillance to provide growers with practical control measures.

D. Variety Evaluation

1. Comparison of growth characteristics of forage-crop species produced in different environments. Vegetative and floral characteristics of birdsfoot trefoil, orchardgrass, and timothy were investigated at Lafayette, Indiana. Pilot seed increases for these studies were made at Shafter and Tehachapi, California; Logan, Utah; and Prosser, Washington, where data were obtained on floral development and on the components of seed yield. Growth characteristics of the progeny population of a 4-clone birdsfoot trefoil selection grown from seed produced at the Western locations were different from those of the progeny population grown from seed produced at Beltsville, Maryland. The progeny populations from seed harvested at Logan, Utah, were more similar to the progeny populations from seed produced at Beltsville. Populations from seed grown at Prosser, Shafter, and Tehachapi differed significantly in floral and growth characteristics from populations grown from seed produced at Beltsville. Similar comparisons were made with seed grown at the same Western locations for a 7-clone synthetic variety from Pennsylvania. Variations among the populations from seed from the different locations were of considerably lesser magnitude.

Growth characteristics of the progeny of the 4-clone Essex timothy variety from seed produced at Prosser, Washington, were considerably different from the characteristics of the seed produced at Ithaca, New York, the location where the four clones were selected. Seed produced at Tehachapi, California, and Logan, Utah, resulted in progeny populations most similar to those derived from seed produced at Ithaca, New York. The progenies of two of the clones grown from seed produced at all three Western locations were similar. The other two clones showed strong location interactions.

Seed of S-48 timothy produced in Minnesota, Canada, and Washington resulted in progeny populations that were similar to the basic seed originally introduced from England. In contrast, seed produced in California and Oregon resulted in plant populations considerably different from those of the basic seed. The most significant observation was that seed representing the first

generation increase in England resulted in plant populations that were more similar to those grown from seed produced in Oregon and California than the original seed or those from seed produced in Minnesota, Canada, and Washington. The primary varietal change noted in the first generation increase was due to earlier flowering plants. The late flowering characteristics of this variety can be readily lost during seed multiplication unless seed is produced in the proper environment.

Progeny population variations reported reflect the strong affect of photoperiod, temperature, and management on the flowering physiology of birds-foot trefoil and timothy. Evaluation of succeeding generations will provide data as to whether the changes were of such magnitude as to affect the performance of the varieties when grown for forage in their regions of adaptation. These data illustrate the precautionary steps which must be taken by the plant breeder to select clones which "nick" in a specific environment in order to produce a reasonably stable synthetic variety. Furthermore, the data indicate the need for more basic information on floral initiation and development and the integration of these with climatic conditions in the primary seed-producing regions.

Seed was produced in seven different States and three generations of increase, representing a wide range of environmental conditions. No differences were noted among the plant populations of 45 seed samples of Potomac orchardgrass, a synthetic variety having 512 parental clones. From these data, it can be concluded that very little genetic change has occurred in Potomac orchardgrass during the generations of increase in the Western States.

2. Influence of environment on population shifts in red clovers. Results obtained under a P. L. 480 research agreement in Finland, terminated in June 1965, indicate only slight population shifts when seed of Finnish white clover and diploid alsike and red clover varieties were produced in the United States and Canada. It was found that the farther north the seed is produced, the smaller the shift in the variety. However, difficulties with seed production in northern climates establish the need for the seed multiplication under the more favorable climatic conditions in the U.S., although there is a slight genetic shift. Evaluation tests indicate commercial increases of these varieties can be made in the U.S. for one or more generations without significant change. Loss of stand due to root rot has generally limited the increase of Finnish clover varieties in the U. S. to one seed harvest following the year of establishment.

3. Influence of environment on genetic shift in forage varieties. Research under a P. L. 480 project in Israel has shown the occurrence of genetic shifts when varieties originating in northern latitudes are grown for seed at 33° latitude. The first generation increases of Iso tetraploid red clover and Alaskland red clover were different in flowering habit from the stocks furnished by the originating breeder. In orchardgrass, there was a higher

percentage of semiprostrate plants in Pennlate than in the original seed. The changes in Tammisto orchardgrass of different genetic origin were negligible. No differences in floral response and flower color were found among alfalfa seed lots produced in Israel. First-generation seed of these varieties has been planted to produce second-generation seed.

4. Determination of genetic quality of West German forage-crop varieties produced in the U.S. A P.L. 480 project has been initiated in Germany to study the effects of temperature, photoperiod, and management on plant ecotypes constituting several grass and legume varieties originating in Germany. The pilot seed increases have been established in both cooperating countries. When two seed crops are harvested, representing the first generation of increase, evaluation trials will be initiated in Germany. In the U.S., the pilot seed increases are made in three different geographic regions.

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WEED AND NEMATODE CONTROL
Crops Research Division, ARS

Problem. Weeds cause losses in crops, orchards, grazing lands, forests, water supplies, and irrigation and drainage systems. These losses can be reduced by finding more effective chemical, biological, mechanical and combination methods of weed control.

Plant-parasitic nematodes occur in all soils used for growing of crops and attack all kinds of plants grown for food, forage, fiber, feed or ornamental purposes. Severity of attack by certain fungi is increased if nematodes are present. Nematodes also have been known to be the vectors of several plant viruses. There is need for improvement in methods of controlling nematodes on grain and forage crops.

USDA AND COOPERATIVE PROGRAM

Much of the weed and nematode control research in the Department is cooperative with State Experiment Stations, other Federal agencies, industry and certain private groups; and is cross commodity in nature. The total Federal weed control program involves 82.0 professional man-years' effort. Of this total, 4.5 man-years are specifically directed to weed control in cereal crops, 1.1 in rice; and 23.5 in forage and range plants. The total Federal nematode control program involves 29.3 professional man-years' effort of which 0.1 are devoted to cereals and 0.7 to forage and range crops. There are P.L. 480 projects at Poznan, Poland, to study the effects of chemical and mechanical control methods on weeds, corn, and associated crops and at Lucknow, India, for biochemical studies on angiosperm parasites--dodder and witchweed; at Orissa, India, for nematodes attacking rice and at Jodhpur, India on nematodes attacking millets.

PROGRAM OF STATE EXPERIMENT STATIONS

The total State scientific effort devoted to weed control research is 110.6 scientist man-years and to nematode identification, physiology and control 53.6 scientist man-years.

PROGRESS - USDA AND COOPERATIVE PROGRAMS

A. Weed Control

1. Small Grains and Sorghum. Studies in Kansas on the use of herbicides to replace part or all tillage operations in a wheat-sorghum-fallow rotation showed that minimum tillage for a period of five years did not adversely affect wheat yields in 1965. Herbicide treatments on wheat stubble in 1964 did not result in complete weed control in sorghum grown in 1965, but when the chemical treatment was supplemented with minimum tillage for effective weed control, sorghum yields were greater than obtained on conventional tillage.

Picloram again gave outstanding control of field bindweed in Kansas in 1965. Although residue studies are incomplete, it appears that picloram may be less persistent in the soil than many of the herbicides commonly used for field bindweed control. For the second season, a mixture of picloram or dicamba with MCPA gave better control of hard-to-kill broadleaf weeds in wheat than MCPA alone. Mixtures of MCPA or 2,4-D with picloram were equally effective. Ioxynil and bromoxynil were about equally effective in controlling broadleaf weeds. In a study on the response of ten wheat varieties to dicamba, tolerance to the herbicide varied considerably. The yield of the most resistant variety was not affected by 1/2 lb/A of dicamba while the yield of the least resistant variety was reduced more than 40% by 1/2 lb/A.

2. Rice. In Arkansas, PCP, molinate, trifluralin, and several newer herbicides incorporated in the soil were as injurious to rice as to weed grasses. Selectivity of these herbicides appeared to be due to differential placement in the soil rather than physiological tolerance.

Bluebonnet 50, a 150-day maturing rice, and Nato (130-day) competed better with barnyardgrass than Vegold (110-day). Weed grasses reduced yields of Bluebonnet 50 gradually, and Vegold rapidly, as the period of competition increased.

Propanil applied continuously for 6 years controlled susceptible weeds, did not injure rice, and was as effective alone as rotating other herbicides with it. Propanil, CIPC, swep, OCS-21944, NPA, amiben, and linuron used continuously and alternately did not damage rice or soybeans grown continuously or alternately. Propanil at 1-1/2 lb/A in two applications or at 1 lb/A in three applications was less effective than propanil at 3 lb/A applied in one application at the recommended time.

3. Grasses Grown for Seed. Seeds of bluegrass and bentgrass may remain viable in soil for extended periods. When raised for seed production, these crops may contaminate future seed crops by volunteer plants coming from seed in the soil. Recommended weed control practices in establishing perennial grasses for seed production effectively reduced the number of volunteer crop plants from seed in the soil.

The sensitivity of creeping red and chewing fescue varieties to recommended herbicides has been an obstacle to effective weed control in the past. In 1965, prometryne applied in October to fields planted in March controlled most of the weed grasses in the first seed crop without reducing yields. Kingstown velvet bentgrass, a new seed crop in western Oregon, appears very sensitive to herbicides used in other bentgrasses. Additional studies are needed to find safe methods of controlling weeds in this species.

Bladder campion was selectively controlled in Newport bentgrass seedfield by spring applications of silvex, dicamba, or picloram. Adding a sticker-spreader to lower rates of these herbicides did not increase their effectiveness on bladder campion.

4. Dodder Control in Alfalfa. Incorporation in the soil of several preemergence herbicides applied for control of dodder reduced their effectiveness. Dodder is a plant which is rootless and apparently dilution of the herbicide through incorporation reduced absorption by the germinating seedling.

5. Weeds in Grazing Lands

Musk Thistle. At Lincoln, Nebraska, musk thistle bloomed in the first week of June in 1965 regardless of the date of planting (13 dates from June to September) in 1964. Musk thistle seeds placed in different pasture types under three different levels of management (continuous grazing; grazing deferred and rotated; and protected from grazing) resulted in the most rosette plants in October in smooth brome grass and intermediate wheatgrass pastures protected from grazing. Plants also became established in the best bluegrass pastures. The smallest number of thistles to become established was in the warm season grass mixture protected from grazing.

Rush Skeletonweed. Individual rush skeletonweed plants disperse 8 to 12 thousand seeds per plant during a single season in Washington. Naturally dispersed seed exhibited no dormancy and had a viability of 95%. Less mature, hand collected seed showed much lower viability.

Larkspur and False Hellebore. In Utah, field results were duplicated in the laboratory to obtain seed germination of larkspur and false hellebore. Seeds of these species, even when buried, were found to either germinate or decompose almost 100% during the first year. Flowering of Duncceap tall larkspur was not influenced by photoperiod during the year the stem produces flowers. In the natural habitat, this species flowers in late June but plants transplanted into the greenhouse, from the same site, produced flower buds in January.

Downy Brome. In 1965, enough seed of downy brome was present in samples of litter and soil in Nevada to have produced 1,200 plants per square foot. Actually, 700 plants grew with 500 viable seeds per square foot remaining in undisturbed areas. Seed production from the plants that grew in 1965 was 800 per square foot which added to the seed reservoir for 1966. On chemical fallow areas, seed production was eliminated the year of atrazine application. The number of seed carried over from the previous year was 500 per square foot, but by October the number of seeds was reduced to 270.

Herbicides Affect Poisonous Characteristics of Weeds. In experiments on the effect of herbicides on the poisonous characteristics of tall larkspur, timber milkvetch, and western false hellebore fed to cattle in Utah, 2,4,5-T and silvex affected toxicity only in timber milkvetch. Timber milkvetch treated with 2,4,5-T was somewhat less toxic to cattle, while plants treated with silvex were much less toxic than control plants.

Multiple Methods of Chemical Analysis Improve Reliability. Research in New York illustrates the errors than can occur if only one method of analysis for herbicides is used. It was found that two natural plant pigments, the all-trans isomers of 3,3'-dihydroxyalphacarotene (lutein) and 5,6,5',6'-diepoxyzeaxanthin (violaxanthin) isolated from brome grass and timothy have retention characteristics identical to 2,4-D on several types of chromatographic columns commonly used in analysis for this herbicide. These properties are exhibited over a wide range of chromatograph parameters. Concentration of the pigments vary with stage of plant development, climatic variables affecting rate of plant growth, and levels of herbicide in plants.

Ecological Studies on Weed Species. In a PL 480 grant with Banaras Hindu University, it was determined that there are 95 species of weeds of agricultural importance which are common to the United States and the Gangetic Plains of India. There is evidence that the frequency of invasion of such weeds from either country to the other has been increasing during the last 200 years.

6. Weed Control in Turf. Siduron controlled crabgrass in a bluegrass lawn for the entire growing season at Beltsville, Maryland, when applied before crabgrass germination in the spring. When siduron was mixed with seed of Kentucky bluegrass, red fescue and Penncross bentgrass, it did not reduce germination or emergence of the seed compared to untreated seed when planted after 9 months in storage. Siduron incorporated $\frac{1}{2}$ inch deep just before planting several Agrostis, Festuca, Lolium, and Poa species, selectively controlled barnyardgrass, crabgrass, seedling nimblewill and fall panicum.

7. Weed Control in Forage Crops. In Maryland, trifluralin incorporated in the soil to a depth of 5 inches prior to planting gave full season control of annual weed grasses in spring-seeded alfalfa, birdsfoot trefoil, and crown vetch without affecting legume nodulation.

In New York, EPTC, as a pre-planting soil-incorporated treatment, also had no adverse effect on legume nodulation. Establishment of birdsfoot trefoil and alfalfa by use of band-subsurface herbicide placement techniques for weed control continued to be effective. EPTC, applied in this manner as a

liquid or impregnated on granular clays or pelleted fertilizers, gave good control of annual weed grasses and many annual broadleaved weeds. Another practice showing some promise was the placement of activated charcoal in a narrow band over a seeded row of legumes at the time of seeding which protected alfalfa from injurious effects of G-36393, an analog of ametryne, applied just after planting.

In New York, paraquat applied to emerged weeds immediately before seeding alfalfa, birdsfoot trefoil, and oats, on a seedbed that had been prepared six weeks earlier in the spring, resulted in season-long control of all annual grass and broadleaved weeds. Establishment of legumes as a result of the nearly perfect pre-planting weed control was outstanding.

8. Weed Control in Pastures and Rangelands. In Nebraska, best control of musk thistle with herbicides was obtained from applications in the full rosette stage or just before bolting in the spring. Several herbicides gave excellent control when applied at these growth stages, with no damage to brome grass. At earlier and later growth stages, picloram and dicamba were more effective than were 2,4-D, 2,4-DB, and 2,4,5-T. Picloram caused considerable injury to brome grass.

Picloram also gave excellent control of whorled milkweed in Nebraska in 1963 and 1965 studies. Plots treated in 1963 with picloram were still free of whorled milkweed in 1965. Silvex, 2,4-D, amitrole, and 2,4,5-T treatments resulted in killing initial topgrowth in both studies. However, the plots treated in 1963 were reinfested the following year. In Mississippi, picloram and 2,4-D gave 100% and 75% control of tall ironweed respectively.

Cattle preferentially grazed broomsedge where nitrogen was applied. Extra growth was mowed at least twice during the season. The stand of broomsedge was reduced. In 3 to 4 years, broomsedge was almost eliminated. After three to four seasons of fertilizing (with nitrogen or complete fertilizer), mowing (twice each season), and grazing broomsedge, dallisgrass stand was greatly increased and broomsedge was reduced. These management practices were as effective as mechanical renovation or herbicide treatments in controlling broomsedge. Nitrogen fertilization on broomsedge pasture increased the protein content of young growth of broomsedge about 50% with and without lime, phosphate, and potash.

Four years of study in Indiana on the effect of Canada thistle density and its control on alfalfa production and utilization have confirmed that continual mowing after each grazing period will give excellent control of this weed. Yields of alfalfa were reduced more than 7-1/4 tons over a 4-year period, by an initial stand of 2 Canada thistle plants per square foot. Vegetation eaten by sheep over the same period was 4-3/4 tons less. This loss is equivalent to 1-1/2 to 2 years additional grazing.

Studies on the control of western false hellebore were continued in Utah. Silvex gave excellent control on dry sites but only fair control on water-logged soils. Picloram controlled false hellebore on all sites and can be used earlier in the season than silvex.

In the Northwest, granular formulations of picloram and dicamba applied in the fall or in the spring, gave excellent control of bracken fern. Granular formulations of both herbicides applied preemergence, particularly in the fall, were superior to foliage applications in the spring. Picloram applied either to the foliage or as a granular application to the soil in the fall was superior to all other herbicides evaluated for control of rush skeletonweed. A mixture of silvex plus picloram showed additive effects and was effective in controlling Dalmatian toadflax in Washington.

In weed control research in Nevada, the effectiveness of chemical treatments on downy brome-infested rangelands was altered principally by timing of treatment, herbicide persistence, broadness of control spectrum, precipitation pattern, germination characteristics of weeds and forage plants, and total reproductive capacity of downy brome and associated broad-leaved weeds. Postemergence weed control reduces competing vegetation in the first growing season and increases establishment of perennial grasses as is also accomplished by spraying paraquat before seeding. Atrazine as a chemical fallow treatment applied the year previous to seeding improves perennial grass establishment by controlling downy brome the seeding year and by conservation of moisture the year prior to seeding.

9. Weed Control in Native Grass Seed Production. Bromacil, and mixtures containing bromacil, were the most effective herbicides in controlling both broadleaved weeds and weed grasses and improving sideoats grama seed production and quality in Nebraska. Other herbicides significantly reduced broadleaved weed yields but were ineffective in controlling the weed grasses. Weed competition where herbicides were ineffective significantly lowered seed production. Excessive rainfall after treatment was probably responsible for decreased weed control and increased crop damage from some of the herbicides. Pyriclor treatments gave good weed control but caused chlorosis in the early growth of treated crops.

B. Nematode Control

1. Forage and Range. The alfalfa stem nematode damaged alfalfa in 18 of 29 counties in Utah. Stem nematode resistance in Kayseri alfalfa was increased from 57% to 70% during 1965. The systemic organo-phosphates, dimethoate, cynem, methyl demeton-R, Bayer 37289, and Bayer 35141, gave good to excellent control of alfalfa stem nematodes in greenhouse tests, and may prove beneficial in areas where climatically suitable resistant

alfalfas are not available. Alfalfa breeding lines UN-25, UN-26, and M-9 have good resistance to the northern root-knot nematode in the temperature range 23-25°C, but at 30°C resistance is reduced but remains better than Lahontan and Ranger. At Tifton, Georgia, several sources of resistance to the peanut, northern, and javanese root-knot nematodes have been found in yellow Lupine 'Weiko III'. Vicia serratifolia, V. conigera, V. calcarata, and 17 breeding lines from V. sativa X V. angustifolia crosses were highly resistant to the southern, cotton, and javanese root-knot nematodes but susceptible to the peanut and northern root-knot nematodes. Twenty-six lines of serica lespedeza were selected for moderate resistance to the northern root-knot nematode in 1964, and in 1965, the progeny showed high homozygous resistance, indicating progression from moderate to high level resistance in two cycles of selection. Of forty-two strains of bermudagrass turf studied, only Tifton 57 and Ormond were selected for further breeding on the basis of superior performance and low populations of sting and stubby-root nematodes. Nematodes on bermudagrass turf were controlled for 7 months with phorate, disulfaton, and Bayer 25141. Diazinon and cynem controlled nematodes for approximately 2 months, which was equal to dibromochloropropane (Nemagon). All chemicals greatly improved root and vegetative growth.

2. Grain. All commonly grown grain sorghum varieties in the South Plains of Texas are highly susceptible to the cotton root-knot nematode, which causes root-damage and reduces sorghum growth, and increases populations of nematodes on subsequent crops. Additional research is needed to develop root-knot resistant grain sorghum, and to evaluate more fully the interrelationships of nematodes and sorghum grain-cotton cropping practices.

A number of plant-parasitic nematodes, including lance and root-lesion nematodes, were associated with damage to millet at Jodhpur, India, under a PL-480 project. A survey of rice at Orissa, India, under a PL-480 project, established thirteen species of plant nematodes can injure rice. The forms causing greatest damage were root-knot, stunt, spiral, rice, and cyst nematodes.

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CORN, SORGHUM AND SMALL GRAIN INSECTS
Entomology Research Division, ARS

Problem. Many species of insects cause losses amounting to millions of dollars annually to corn, sorghum, and small grains. It is estimated that 25 species of insects cause an annual loss of \$900 million to corn alone. The European corn borer and corn earworm are two of the most destructive insects in the country, and corn rootworms are serious pests of corn. Armyworms attack corn and small grains. In certain years the greenbug causes widespread losses to wheat, barley, and oats in the Central and Southeastern States, and the Hessian fly and wheat stem sawfly annually damage the wheat crop in certain areas. The cereal leaf beetle, first identified in the United States in 1962 from Berrien County, Mich., now occurs in many counties in Michigan, Indiana, and Ohio, and is a threat of unknown proportion to small grain crops. Such examples of the destructiveness of insects to corn, sorghum, and small grains point up the need for extensive research that will lead to the development of adequate means for the control of these important crop pests. Progress has been made toward the solution of some of the insect problems encountered in the production of grain crops but more effective, more economical, and safer insect control measures are needed. Research is essential to find insecticides that can be applied to grain crops, that will not leave residues harmful to animals consuming the feed, that will not be a hazard in milk, and meat, and that will not be detrimental to beneficial insects or to fish and wildlife. The appearance of resistance to certain insecticides in several grain insect pests stresses the need for basic information to overcome this problem. Additional emphasis should be placed on research to develop crop varieties resistant to insects and on biological and cultural control methods. New approaches to insect control, such as sterilization techniques and attractants, require expanded investigation. Research is also needed on insect vectors and the role they play in the dissemination of important plant diseases. The heavy losses in oats, wheat, and barley due to barley yellow dwarf virus, and in corn due to maize dwarf mosaic and corn stunt recently found in several North Central and Southern States, indicate the importance of research in this field.

USDA AND COOPERATIVE PROGRAM

The Department's program involves both basic and applied research directed toward developing more efficient control methods for insects attacking grain. All studies are conducted in cooperation with State Experiment Stations in the several States where research is underway. Studies on evaluating and developing varieties of grain which resist insect attack are conducted in cooperation with State and Federal agronomists and plant breeders and research on insect transmission of diseases of grain crops is in cooperation with State and Federal plant pathologists. This research includes studies on Hessian fly at Lafayette, Ind., and Manhattan, Kans.; cereal leaf beetle at Lafayette, Ind., and East Lansing, Mich.; aphids and

mites attacking small grains at Stillwater, Okla., Brookings, S. Dak., and Tifton, Ga.; wheat stem sawfly at Fargo, N. Dak., and Bozeman, Mont.; corn earworm at Tifton, Ga., State College, Miss., and Lafayette, Ind.; fall armyworm, pink scavenger caterpillar, and rice weevil at State College, Miss., and Tifton, Ga.; soil insects attacking corn at Brookings, S. Dak., State College, Miss., and Tifton, Ga.; corn leaf aphid at Brookings, S. Dak.; southwestern corn borer at Stillwater, Okla., and State College, Miss.; European corn borer at Ankeny, Iowa, State College, Miss., and Wooster, Ohio; corn earworm, sorghum midge, sorghum webworm, and corn leaf aphid on sorghums at Stillwater, Okla., and Tifton, Ga.; and insect transmission of grain diseases at Manhattan, Kans., State College, Miss., and Brookings, S. Dak. Research to evaluate improved equipment for application of insecticides to grain crops is underway at Ankeny, Iowa, and Tifton, Ga., in cooperation with Federal agricultural engineers. Work on corn rootworms is being conducted at Brookings, S. Dak. Additional research is being conducted under ARS contracts and grants on the biology and control of the cereal leaf beetle with Michigan, Indiana, and Ohio Experiment Stations, soil insects attacking corn with the University of Nebraska, and vectors of corn stunt virus with Mississippi State College, nature of resistance of corn to the European corn borer with Iowa State University, and insect communication in the infrared region with Michigan University, Ann Arbor, Mich., transmission of viruses causing stunting of corn, with Missouri University, ecological factors affecting efficiency of Trichogramma spp. with Louisiana State University.

The Federal scientific effort devoted to research in this area totals 42.3 scientist man-years. Of this number 10.8 is devoted to basic biology, physiology, and nutrition; 3.5 to insecticidal and cultural control; 1.8 to insecticide residue determinations; 4.1 to biological control; 2.9 to insect sterility, attractants and other new approaches to control; .5 to evaluation of equipment for insect detection and control; 15.7 to varietal evaluation for insect resistance; 1.8 to insect vectors of diseases; and 1.4 to program leadership.

Certain phases of this research are contributing to regional research project NC-20 "Factors Influencing European Corn Borer Populations." A P.L. 480 project, E8-ENT-1, "Population Dynamic Studies on Calligypona pellucida (F.) and the Nature of Injuries Caused by this and Other Leafhopper Species (Fulgoridae) on Cereals, Especially Oats and Spring Wheat" is underway at the Agricultural Research Centre, Department of Pest Investigation, Helsinki, Finland. Another P.L. 480 project, A10-ENT-5, "Host Plant-Vector and Host Plant-Virus Relationships of Rough Dwarf Virus of Corn and Methods for Control of the Disease" is being conducted at the Hebrew University, Rehovoth, Israel. A7-ENT-25 in India is concerned with "Research on Insect Pests of Maize with Special Reference to Stalk Borers." Projects A7-ENT-31 in India, "A Study of the Host Plant-Vectors and Host Plant-Virus Relationships in the Rough Dwarf Virus Disease of Maize, in Order to Develop Efficient Methods for the Control of the Disease," and E21-ENT-14, in Poland, "Studies Regarding the Bionomics, Economic Importance and Natural Control Factors Affecting Oulema Species/Cereal Leaf Beetle in Poland," and E30-ENT-3 in Yugoslavia on "Parasites, Predators, and Pathogenic Organisms Study of the Cereal Leaf

Beetle, Resistance Investigation of Domestic and Foreign Small Grain Varieties to Oulema melanopa," are also being conducted.

PROGRAM OF STATE EXPERIMENT STATIONS

A total of 39.2 professional man-years is devoted to this area of research.

PROGRESS -- USDA AND COOPERATIVE PROGRAMS

A. Basic Biology, Physiology, and Nutrition

1. Corn Insects. Average seasonal populations of the western corn rootworm at Brookings, S.Dak., dropped to a low of 1.0 adults per 100 plants in 1965. In contrast northern corn rootworm populations increased sharply to 522.3 adults per 100 plants for the season. Similar changes in the northern and western corn rootworm adult population ratios have been reported in State survey reports from Minnesota and Iowa.

Laboratory and field studies have shown that contrary to common believe the corn rootworms can use plants other than corn for larval development. Tests in field cages indicated that green and yellow foxtail grass, Minter wheat, Omugi barley, Oahe intermediate wheatgrass, in addition to corn supported rootworm development to the adult stage. None supported as large a population as corn. Adult western corn rootworms produced viable eggs when reared as larvae on wheat, barley, and green foxtail grass. Northern corn rootworm adults laid viable eggs when they were reared as larvae on wheat and the southern corn rootworm produced viable eggs when reared on wheat, barley, and yellow foxtail grass.

Corn plots with an average of less than 1% of the plants damaged by corn rootworms had a yield of 115 bushels per acre. Plots with 60% of the plants showing damage had a yield of 88 bushels per acre, or a 23% yield loss due to corn rootworm.

Laboratory studies indicate that fertile backcrosses are possible with parental stocks. F_3 hybrid beetles ($P_1 = \text{WCR} \sigma \times \text{NCR} \text{♀}$) were backcrossed to northern (NCR) and western (WCR) corn rootworms. Eggs collected from the 4 reciprocal crosses were incubated and only the eggs from F_3 hybrid $\text{♀} \times \text{NCR} \sigma$ failed to hatch. Adults were reared from eggs laid by F_3 hybrid $\text{♀} \times \text{WCR} \sigma$, $\text{WCR} \text{♀} \times F_3$ hybrid σ , and from $\text{NCR} \text{♀} \times F_3$ hybrid σ . All F_4 adults appear morphologically similar to WCR.

Ecological data on the corn borer population in Boone County, Iowa, showed an early spring population of 3,417 borers per acre, midsummer borer population of 2,538 borers per acre, second brood pre-harvest population, 4,313 borers per acre, and after harvest survey of borers going into hibernation of 1,625 borers per acre. Light trap catches at Ankeny Farm indicated two complete generations of corn borers with a partial third.

Field experiment investigations on the biotypes of the European corn borer indicated incidence of diapause among the F₁ generations was the best criterion for separating one biotype from another. Borer cultures from Minnesota had a higher percent diapause than those from Missouri, with those from Iowa intermediate between the two. No biotypes capable of damaging previously resistant crop varieties were found.

Tests conducted at Ankeny, Iowa, show that pollen shedding at time of infestation had a profound effect in increasing second brood larval establishment on susceptible inbreds, but very little or no effect on resistant inbreds.

Greenhouse studies show that 1/4 to 1/3 of the corn borer larvae hatching from eggs laid on the corn plant migrate off the plant. No differential in larval migration from susceptible inbred WF9 and resistant inbred C.I.31A was observed. There was a wide differential in larval survival after egg hatch on susceptible and resistant inbreds.

Diet improvement studies provided a laboratory rearing diet which is an improvement over the diet used for the past several years. Wheat germ has been substituted for ground corn leaf, and changes made in vitamin and salt content. Larvae reared on the new diet are more uniform in size and require less of it for complete development.

A study of spermiogenesis in corn borer larvae showed that testis volume increases throughout larval and early pupal life, then with emergence of the adult insect, it gradually decreases. The testis of third instar larvae contains essentially only spermatogonial and primary spermatocytal cysts; secondary spermatocytal cysts predominate in the 4th instar larva; spermalidal cysts are at peak numbers in early pupal life; spermatozoa appear in full grown larvae and predominate in late pupal testes; when the insect reaches adulthood it contains its full complement of sperm.

Chromic oxide, added to the dry matter of test diets fed to European corn borer larvae, provided colorimetric method for measuring diet consumption and utilization during growth. These tests showed that growing corn borer larvae may consume up to 40 mg of food per day and utilize as much as 52% of it. The larvae utilize their food most effectively during the 3rd and 4th instars - first and 5th instar larvae being the least efficient.

In Mississippi in 1965, 70% of the ears in all corn fields were infested with the corn earworm, 89% of the ears were infested with pink scavenger caterpillar, and 18.6% with rice weevil. Southwestern corn borer damage to dent corn in Mississippi continues to be a serious problem. The percentage fields infested increased from 66.2% in 1964 to 78.4% in 1965. The southwestern corn borer spread to 2 additional uninfested counties in Mississippi, 2 in Alabama, and 2 in Tennessee.

At Tifton, Ga., a commercially available food packaging machine was modified to mass rear the fall armyworm. The machine fills 1 ounce plastic containers with artificial diet, dispenses a larva on the diet, and caps the container in one continuous operation. The rate may be varied between 2,000 and 4,500 containers per hour. One thousand pupae can be produced with this machine at a cost of \$14.60, adults for \$16.50, and larvae \$9.20.

Experiments at Tifton indicate that the simplex in the male fall armyworm reproductive system may be used to determine if the males had mated. In the unmated condition the simplex is filled with a substance of dark coloration, whereas in the mated condition the simplex is transparent.

At Tifton tests indicated that mating in the corn earworm increased when the surface of the insect cages was lined with low emissivity aluminum foil and when a corn plant was added to the cages. Highest mating was obtained when both factors were combined (83.5%). Higher mating was found in foil-lined cages without plants than was found in unlined cages with plants.

Tests conducted at Tifton, Ga., showed that near infrared had a highly stimulating effect on flight of the Indian meal moth. After the moth's eyes had been day conditioned ("gated") by a low intensity 5200 Å visible light source, the moths flew continuously until death - at 48 hours. When infrared was used in conjunction with light of the ultraviolet region, infrared had no stimulating effect on moth flight.

2. Small Grain and Sorghum Insects. Surveys conducted during the 1965-66 crop year indicate approximately 10 million acres of Hessian fly resistant varieties, developed through the cooperative efforts of USDA and Experiment Station personnel, were grown in more than 24 States. This represents an increase over last year of 1-1/2 million acres.

A new Hessian fly resistant soft red winter wheat, Benhur, was developed and released in Indiana. This wheat, besides being resistant to the major wheat diseases, is resistant to the new race of Hessian fly (Race B) threatening the wheat crop in southern Indiana. Benhur covers the H_C gene for resistance and protects the wheat from Races A and B.

Hessian fly populations are building up in susceptible wheat varieties grown in Michigan. Samples from 111 certified wheat fields in Michigan showed 82% of the fields to be infested with Hessian fly with an average infestation of 16.9% for those wheats having no Hessian fly resistance, and only 2.6% for those wheats having fly resistance. Hessian fly resistant wheats were slightly infested in those counties bordering Indiana, due to a Race B buildup resulting from large acreages of Monon wheat being grown in the area.

Investigations conducted at East Lansing, Mich., indicate that oviposition and viability of eggs of the cereal leaf beetle might be improved by use of a shorter cage with a higher relative humidity and fewer insects per cage.

These studies also indicate that a sex ratio weighted possibly as high as 4:1 in favor of the female will produce a greater number of eggs. In several instances females have laid up to 400 eggs each and in one case one female had laid over 900 eggs in a five-month period.

Studies on the life stages of the cereal leaf beetle indicate that the two main factors affecting reproductive activity of the cereal leaf beetle are temperature and photoperiod. Optimum conditions for the maximum reproduction in this insect are 80° F, 16 hours of light, and 50-70% relative humidity. Beetles raised under 12, 20, and 24-hour light periods failed to produce eggs. Any group of newly-emerged adults held under optimum conditions is capable of egg production after a period of 25 to 30 days. An individual female is capable of producing up to 1,251 eggs during her life span and up to 28 eggs a day.

An assay method suitable for determining degrees of induction or termination of diapause in the cereal leaf beetle was developed. Serial sections of the brain and related glands were made of pre-, post-, and diapause beetles, both male and female, and stained, using paraldehyde fuchsin neural-secretory techniques. Differences were observed between pre- and diapause beetles versus post-diapause beetles. Both median neural-secretory cells and the corpora allata appeared to be active in secretory granules in the post-diapause condition.

Sorghum midge infestations in Oklahoma and the Plainview-Lubbock Area of Texas were light in 1965 - causing little or no damage. Observations have indicated that sorghum planted at "normal" planting dates and blooming in late July or early August, will escape damage from the midge.

In greenhouse tests at Stillwater, Okla., heavy infestations of the English grain aphid caused plant stunting and weight losses, ranging from 58% in Will barley to 89% in Cimarron oats.

Rearing tests conducted in plant growth chambers having day-night high-low temperature values of 60-35, 70-45, 80-55, and 90-65 degrees F, respectively, indicated that length of the reproductive period in days was inversely proportional to the temperatures. Mean temperatures of 50, 40, 36, and 34° F, progenies of 12 greenbugs in each chamber were 263, 559, 737, and 886. Reproduction began 27, 12, 9, and 8 days after start of the experiment in the respective temperature programmed chambers.

In tests conducted at Brookings, S. Dak., color had a definite effect on reproduction and survival of cereal aphids. The four most common cereal aphids in South Dakota, Macrosiphum avenae, Rhopalosiphum padi, R. maidis, and Schizaphis graminum were fed artificially through Parafilm membranes under different colored filters for a period of 10 days. Responses to specific colors differed with species, but in general green and orange filters resulted in most favorable survival and reproduction. It was also shown that variable light intensity was not involved in aphid response to

colored filters. Color preference tests with these same aphids indicated that adult apterae and nymphs of R. padi and nymphs of R. maidis preferred green over yellow, red, or blue. Apterae of R. maidis and apterae and nymphs of M. avenae and S. graminum preferred yellow.

Five species of false wireworms, Eleodes opacus, E. hispilabris, E. extricata, and Embaphian muricatum, and Eleodes suturalis, have been successfully reared in the laboratory at Brookings, S. Dak. Seeds most preferred for food were alfalfa, hullers barley, flax, forage sorghum, hullers oats, rye, and wheat.

A portable, self-powered, vacuum insect collector was used to collect adult sawflies in a heavily infested field of wheat at Minot, N. Dak. Adults collected were used to supplement field infested wheats in experimental plots. Sawflies collected by this method were subjected to an air flow of high velocity but little adult damage was observed.

In studies conducted under a grant at Purdue University on the behavior of the cereal leaf beetle as affected by climatic factors, it was found that the early summer and fall dormancy of the adult beetle can be interrupted by subjecting the beetles to high sublethal temperature. This treatment has resulted in good production of viable eggs from adult seasonal forms that normally will not produce eggs.

Studies were conducted under P.L. 480 project, E21-ENT-14, Poland, to develop survey methods for measuring distribution and damage of the cereal leaf beetle.

3. Other Insects. Aedes aegypti, when released in a cross-box composed of five compartments, preferred to rest on a surface that was highly infrared reflective.

When released in a room with two CDC miniature light traps, one radiating white light and the other infrared, the majority of mosquitoes collected were taken in the infrared trap. When released in total darkness, the mosquitoes were attracted to an infrared trap, indicating an ability to locate an infrared source. Blood-fed females and mated mosquitoes of both sexes appeared less attracted to infrared than when unmated or honey-fed.

B. Insecticidal and Cultural Control

1. Corn Insects. Twenty-two insecticides in granular formulation were field-tested against first-generation European corn borer larvae, and 23 were tested against second-generation borers at Ankeny, Iowa. Niagara NIA-10242 was an outstanding compound for control. Eleven gave control equal to or superior to DDT in these tests.

Spray formulations, field tested, consisted of 9 and 8 insecticides for first and second generation tests, respectively. Niagara NIA-10242 was

again outstanding for control. Azodrin, American Cyanamid CL-47470, and endrin were equal to or superior to DDT in these tests.

American Cyanamid CL-47470, applied to the soil at time of planting as a systemic insecticide gave satisfactory control for 30 to 40 days. Low volume application of malathion by air gave no control.

In Mississippi two weekly applications of Niagara NIA-10242 at 1.0 pound per acre for second generation southwestern corn borer and two for third generation gave 73.3% control.

At Tifton, experimental chemicals were evaluated in the field for control of the corn earworm and the fall armyworm attacking sweet corn. Five applications of each material was applied during ear development. Insect control was compared to that obtained with a DDT standard. Of the chemicals tested, only Azodrin was significantly better than DDT. Niagara NIA-10242, Mobil M-CA-600, and Shell SD-7438 gave control equal to the DDT standard.

Tests conducted on varieties of sweet corn, with varying degrees of corn earworm resistance, indicated that very little additional control could be obtained by applying DDT to varieties that were resistant to corn earworm attack. A significant increase in control was obtained by applying DDT to susceptible varieties.

The effect of crop removal on oviposition of the northern and western corn rootworms was investigated at Yankton, S. Dak. Corn rootworm egg laying patterns were determined by egg and larval samples, larval damage, and yield loss. Treatments where corn was removed as silage by September 27 showed very little rootworm damage. Where corn was not removed, severe lodging occurred the following year and yield was reduced 23%.

The effect of fall or spring plowing on corn rootworm populations was evaluated. No significant differences were found in plant height or plant population due to plowing treatments. Significantly more corn rootworm larvae and damage were noted in the fall-plowed treatment. No differences in yield were found, indicating that the rootworm damage tended to cancel out the expected yield advantage following fall plowing.

An aldrin resistant strain of southern corn rootworm has been isolated in the laboratory at Brookings, S. Dak., by insecticide treatment of a susceptible strain. The survivors of this treatment are being maintained as a separate culture. Preliminary data indicate that aldrin resistance in this insect could be a single gene recessive character.

2. Small Grain Insects. Tests were continued at Stillwater, Okla., to study the phytotoxic effect of insecticides on sorghums. Methyl parathion and Bidrin applied to 12 grain sorghum hybrids and breeding lines as a spray caused considerable burning. Ethion, diazinon, and carbaryl wettable powder caused no injury. Hybrids RS 610 and RS 626 and entries y-15 and y-16 (both having yellow endosperm) were not injured by any of the insecticides.

Exploratory tests indicate that it is possible to test grain sorghums for insecticide phytotoxicity in a greenhouse at an early stage of plant growth and in a shorter period of time than is required in the field. Microscope slides of leaf sections made from phytotoxicity-susceptible and resistant plants showed the effects of the phytotoxic action in the former. Damaged plants showed (1) disintegration of the chloroplasts, (2) clogging of xylem and phloem tissues (clogging material not yet identified), and (3) shrinkage of the cell walls. In Bidrin-sprayed plants the larger vascular bundles are affected first, later, the smaller ones. The reaction is just the opposite for plants sprayed with methyl parathion. There was no phytotoxicity when plants were sprayed with diazinon.

In Montana oxydemetonmethyl as a foliar spray applied at the rate of 2 pounds per acre with an additive, dimethyl sulfoxide gave 97% control of the wheat stem sawfly. Heptachlor applied as a foliar spray gave 56% control, and oxydemetonmethyl without sulfoxide 13%.

C. Insecticide Residue Determinations

1. Corn Insects. At Tifton rainfall produced a marked reduction in DDT residues on sweet corn. When one inch of artificial rain was applied at the rate of 0.5 inch and 2 inches per hour there was a 34% and 45% decline, respectively, in DDT residue. A second inch applied at the same rates decreased the deposits another 30% and 25%, respectively.

Gas chromatographic methods of analyzing for the chemosterilants, tepa, metepa, methiotepa, hempa, and apholate, were developed by chemists at Tifton, Ga. The methods employ the Malpar flame photometric detector and are sensitive to about 0.1 nanogram.

Studies were conducted at Tifton on the persistence of the chemosterilant, tepa, in fall armyworm moths. Both sexes were fed 0.3% tepa in 10% aqueous sucrose. After ingesting up to 100 μ g to tepa per moth the insects were held in the laboratory under conditions of controlled temperature and humidity. Within 24 hours over 90% of the tepa had disappeared from the moths; over 95% was gone within 48 hours. Only 1 μ g of tepa per insect was recovered from the feces after 48 hours.

Chemists at Tifton have developed gas chromatographic techniques for analyzing residues of Azodrin and Bidrin in raw extracts of sweet corn using a Melpar flame photometric detector. Residues as low as 2 ppb were detected without appreciable interference from the corn extract. A technique was also developed for analyzing residues of Imidoxan and Imidan in corn silage and in sweet corn at levels of 2 ppb for Imidan and 4 ppb for Imidozan.

D. Biological Control

1. Corn Insects. The microbial insecticide, Bacillus thuringiensis, in capsule, granule, and emulsifiable formulations gave excellent control of the European corn borer at Ankeny, Iowa. Laboratory studies with the bacterium showed that both the spore and crystalline inclusion are necessary to kill the borers. These also showed that a viable spore count is not sufficient standardization on which to base a "control" recommendation.

Further evidence of the importance of the borer infecting protozoan, Perezia pyraustae, in corn borer population reductions was noted in Boone County, Iowa, this spring. Field collections of borers showed an incidence of 50% infection at the mid-March survey and dropped to 16% at the May 1 survey. During this same period the borer population dropped from approximately 3,000 larvae per acre to 900 larvae per acre, indicating that the infected borers were less able to survive the climatic and agronomic changes which occurred during this 6-week period.

At Tifton, Ga., 5 generations of the egg parasite, Trichogramma, were reared on fall armyworm eggs sterilized with tepa without any decline in the parasite culture.

Additional information was obtained on the life history of Drino munda, introduced from India, and previously reported as Drino imbrebis, a tachinid parasite of the corn earworm. D. munda can successfully parasitize both the corn earworm and the fall armyworm. However, it requires almost 4 days longer to complete its life cycle on the fall armyworm than on the corn earworm.

Preliminary results on the effects of temperature and dosage on the pathogenicity of the fall armyworm nuclear-polyhedrosis virus indicated an increase in total mortality in the larvae held at 22-25° C. rather than higher temperatures.

2. Small Grain and Sorghum Insects. Field releases involving approximately 5,000 adult cereal leaf beetle egg parasites, Anaphes sp. have been made in Michigan. Recovery of parasites has been successful with approximately 30% of the eggs examined being parasitized.

Five hundred adults of the wheat stem sawfly parasite, Collyria calcitrator were released at Conrad, Mont., in 1965. One adult was recovered in 1966. Field observations and rearing of insects from collected stubble indicates that populations of the native parasite, Bracon lissogaster are building up slowly in cultivated wheat in Montana.

Tests conducted on parasite activity against the wheat stem sawfly at Fargo, N. Dak., showed that susceptible wheat varieties had the highest number of sawflies parasitized by B. cephi. Similarly, higher sawfly

infestation levels had higher numbers of parasitized sawflies. Sawfly larvae in plots of both susceptible and resistant varieties with low infestation levels were also parasitized indicating this parasite has a keen host-searching ability. These tests provided results which support the conclusion that since the number of parasitized sawfly larvae is a function of the number of sawfly larvae in a given area, the use of a wheat variety highly sawfly resistant would reduce parasite numbers materially.

E. Insect Sterility, Attractants, and Other New Approaches to Control

1. Corn Insects. At Tifton, Ga., experimental chemosterilant compounds, ENT 51253, 50990-a, and 51028-a, were equally effective as tepa in sterilizing the fall armyworm moth. However, sterile males produced with chemosterilants were not as competitive for females as untreated males; competitiveness being reduced by as much as 45% in moths sterilized with 45 µg. One hundred percent sterility was achieved when males or females of fall armyworm and corn borer moths were forced to walk over a felt pad saturated with a 10% sucrose solution of 0.3 to 0.15% tepa. This technique for sterilizing moths will be used in conjunction with light traps to trap, sterilize, and release moths back into their wild environments.

Sex pheromones of fall armyworm, Spodoptera frugiperda, corn earworm, Heliothis zea, and true armyworm, Pseudaletia unipuncta, have been found within the last abdominal segment of virgin, laboratory-reared female moths. Purified extracts of fall armyworm and of corn earworm elicited typical sexual reactivity in the virgin male of 3 species of looper, i.e., Trichoplusia ni, Pseudoplusia includens, and Rachiplusia ou. There is some possibility that this activity is due to the presence of impurities.

An insect feeding device utilizing a felt wick impregnated with the chemosterilant, tepa 0.15% and aqueous sucrose 2% has been developed at Tifton, Ga., for use in conjunction with light traps to sterilize moths in their natural habitat. Analysis for tepa at 1, 2, 3, and 5 day intervals indicate that amount of tepa on the wick diminished 20% during the first 24 hours, 53% by the end of the second day, and 86% by the end of the third day.

Preliminary tests indicated that the chemosterilant, apholate, was more effective in sterilizing the southern corn rootworm than metepa or tepa. Females treated with 50 ppm apholate mated with untreated males laid 281 eggs with 38% hatch; untreated females with treated males, 140 eggs with a 4% hatch; both sexes treated 55 eggs with 1.2% hatch. Beetles treated with 100 ppm apholate averaged fewer eggs per female.

Studies on the irradiation of diapausing corn borer larvae indicate that 4000 or 5000 roentgens will sterilize corn borer males, and reduce female egg production and hatch to near zero. Irradiated larvae pupated normally and emerged as apparently normal moths.

A procedure for purification of the sex pheromone of the female European corn borer adult has been developed at Ankeny, Iowa. Purification procedures involved extraction of the substance from the female moth with 1,2-dichloroethane followed by precipitation of inactive lipids at -70°C in solvents of increasing polarity, followed by column chromatography on silica gel and final purification by gas-liquid chromatography. Sixty thousand female moths are now being processed in an effort to obtain a sample of the sex pheromone of sufficient size to allow chemical characterization of the compound.

Male sterilization of the European corn borer was accomplished by feeding 7-day-old larvae tepa-treated media. Eight other compounds representing the different chemicals that have shown promise in other sterilization programs were evaluated, but were not effective.

In studies conducted under a grant at Ohio State University on microbiology and pathologies of Oulema melanopa, it was found the fat body structure of immature developmental stages can be used as a guide for detecting and recognizing new viruses infecting Coleoptera.

2. Small Grain Insects. Apholate was evaluated as a chemosterilant for the cereal leaf beetles. Concentrations above 0.25% suppressed oviposition, and caused mortality of 80%. At 0.10% concentration, oviposition did not occur but mortality was reduced. Concentrations of 0.05% did not prevent oviposition and eggs were not viable. Eggs treated with lower apholate concentrations showed varying degrees of viability.

Eggs of the cereal leaf beetle treated with 1,000, 2,000, 4,000, 8,000, and 16,000 rads. failed to hatch. Hatching, larval survival, pupation, and adult development were normal when given a 1,000 rad. treatment just prior to hatching.

When newly emerged and third and fourth instar larvae of the cereal leaf beetle were irradiated, all levels of treatment above 1,000 rads. resulted in high larval mortality in all cases. At the 1,000 rad. level, there was some mortality but not significantly different from the controls.

Adult emergence of cereal leaf beetle was significantly decreased by irradiation with 1,000, 2,000, 4,000, 8,000, and 16,000 rads. One hundred percent mortality occurred at the 16,000 rad. level.

Four hundred and thirty-one chemical lures were evaluated as attractants for the cereal leaf beetle under field conditions. Twenty-five caused a response in the beetle. During the winter of 1965-66 an additional 200-300 synthetic materials were evaluated under laboratory conditions. None of the lures elicited a response.

Plant extracts prepared from seedlings of barley, oats, wheat, and corn during 1965 using various combinations of solvents and preparation

techniques failed to elicit a detectable response when exposed to adult cereal leaf beetles. Exposure of various populations of adult cereal leaf beetles in field cages during 1966 spring-adult field tests again has failed to indicate any presence of a pheromone.

An olfactometer suitable for testing attractants and repellants on soil insects was developed. The use of this device revealed that a chemical(s) produced by germinating wheat attracts larvae of Eleodes suturalis.

Studies on the effects of gamma-ray irradiation of non-diapausing wheat stem sawfly larvae in wheat stubble continued at Fargo, N. Dak. Sawfly larvae were exposed to total doses of 1,000, 2,000, 3,000, 4,000, and 5,000 rad from a Cobalt 60 source. Adult sawfly emergence at radiation levels of 1,000 and 2,000 rad was comparable to that of the non-irradiated samples. Adult emergence was greatly reduced at the 3,000, 4,000, or 5,000 rad levels.

F. Evaluation of Equipment for Insect Detection and Control

1. Corn Insects. At Tifton, Ga., conventional insecticides applied every day gave better corn earworm control than insecticides applied every other day or every third day. Applications on an alternate day schedule were significantly better than applications made every third day.

Agricultural engineers and entomologists at Tifton have developed a tractor-mounted system for applying insecticides, either as technical or concentrated formulation, at extremely low volumes. One system is capable of applying the toxicant at rates as low as 1 pint per acre. The other system will apply toxicants as low as 1 quart per acre.

At Brookings, S. Dak., colored neon trap lights set up in a corn field indicated that certain shades of red and blue were more attractive to both western and northern corn rootworms than were others. However, corn was more attractive to the insects than were the lights. It was noted that while the sex ratio for both species was about 50:50, 97.6% of the western and 90.1% of the northern corn rootworms attracted to the lights were males.

G. Varietal Evaluation for Insect Resistance

1. Corn Insects. A cyclic hydroxamate (DIMBOA), a constituent of corn tissues, was found to be a chemical factor in the resistance of corn to the European corn borer. Resistant lines of corn produce 10 times more DIMBOA than do susceptible. In bioassay tests the compound inhibited larval development and caused a 50% mortality. Surviving larvae complete development to the adult form with no apparent morphological abnormalities. The biological activity of DIMBOA is sufficient to explain a large degree of the resistance expressed in the field by borer-resistant inbred lines of corn.

Conclusive spectroscopic evidence has shown that a second cyclic hydroxamate is to be found in corn tissue. Structurally, the second cyclic hydroxamate is very similar to DIMBOA and it is expected that the substance will possess biological properties similar to DIMBOA. Analyses have shown that this newly found compound exists at higher concentrations in the more resistant lines of corn.

Tests conducted at Ankeny, Iowa, with a group of 36 hybrids, some new releases and some old, indicated that factors other than leaf feeding resistance, i.e., sheath or stalk resistance and tolerance, are effective in some hybrids in reducing yield losses because of first-brood corn borer stress.

Genetic studies on the use of reciprocal translocations to determine chromosome arms involved in resistance of C.I.31A and B49 to a first brood infestation shows that C.I.31A carries genes for resistance on the short arms of chromosomes 1, 2, and 4, and in the long arms of chromosomes 4 and 6. B49 carries genes for resistance on the short arms of chromosomes 1, 2, and 4, and on the long arms of chromosomes 4, 6, and 8.

In tests at Wooster, Ohio, 8 of 18 inbred lines that indicated resistance or tolerance to the newly discovered virus maize dwarf mosaic also indicated some degree of resistance to a first-brood corn borer infestation.

Inbred GE 315 was the most resistant to southwestern corn borer of 5 inbred lines tested in all possible single cross combinations at State College, Miss. In another single cross test involving 8 inbred lines, Mp315 and Mp337 showed significant resistance to southwestern corn borer.

Resistance studies at Tifton, Ga., indicate that the husk, silks, and kernels all contribute to the amount of feeding (resistance) in an inbred to corn earworm. The factor(s) involved varies with each inbred. For example, inbred 166 had good husk resistance, inbred 245 silk resistance; inbred F-6, husk, silk, and kernels resistance. Pollination of silks increased larval survival.

At Tifton, Ga., 32 corn lines from South and Central America and 20 sweet corn lines were screened in the greenhouse for leaf damage by fall armyworm larvae. Lines showing some resistance were Zapalote Chico, Zapalote Grande, and Antiqua 2-D. In general, sweet corn showed more resistance than dents but further tests are needed to confirm this.

Studies were conducted on the utilization of diets by third instar fall armyworm and corn earworm larvae by the use of chromic oxide as an index compound. In general, the fall armyworm's ability to utilize diets with corn leaves was slightly better than the ability of the corn earworm. While the reverse was true with kernel diets, kernel diets were more highly utilized than silks by both species of insects.

Significant differences were found among 18 lines of corn (leaves, kernels, and silks) in the degree of stimulation to feed they furnished larvae of both fall armyworm and corn earworm. The plant parts that elicited a response as an arrestant in earworm larvae are listed in descending order, as follows: kernels, silks, dried seed, and leaves. In fall armyworm larvae the order was as follows: silks, kernels, leaves, and seed. Kernels with the highest amylose content gave the best feeding stimulant response for both corn earworm and fall armyworm.

In varietal evaluation tests conducted to locate new corn rootworm resistant germ plasm at Brookings, S. Dak., 44 synthetics, 4 Corn Belt inbreds (A297, OH05, OH45B, and 153RM) and 15 inbreds developed by the South Dakota State University, Plant Pathology Department for resistance to root rot also showed resistance to the western corn rootworm. One hundred superior performing inbreds in the breeding program have been advanced to the S-2 generation for further study and potential line development.

In studies conducted at Iowa State University under a grant on the biochemical basis for resistance of maize to attack by the European corn borer, a technique was developed using C¹⁴ to detect both DIMBOA and DIBOA, the chemicals believed to be associated with corn borer resistance in the corn plant. A technique was also developed using the isotope method for the estimating the amount of DIMBOA in corn.

Studies conducted under P.L. 480 project A7-ENT-25, India, which covers research of insect pests of maize, indicate that the stem borer, Chilo zonellus, reduces maize yield by 20%. It was also found that the percentage of infestation and damage was greatest in late sown crops. Several varieties were found to be more resistant to damage than others. The most resistant being Al x Antiqua Gr.I and Cuba 11J.

2. Small Grain and Sorghum Insects. Years of cooperative research between Federal and State agencies in North Dakota culminated in the release of Fortuna, the first hard-red spring wheat variety ever developed that has both sawfly and rust resistant characteristics. Fortuna has out-yielded and been heavier in test weight than the sawfly resistant varieties Chinook and Rescue.

In Michigan the screening for resistance to the cereal leaf beetle in small grain varieties, breeder's lines, introductions, and segregating populations of crosses involving resistance was continued at two locations with approximately 8,000 entries in each nursery. Under the pressure of a high beetle infestation, approximately 55 varieties of winter wheat and a comparable number of spring wheats exhibited resistant reactions. The most common type of resistance observed in the wheats was non-preference of resistant varieties for oviposition. CI's 6671 and 6469 were the most resistant barleys to the cereal leaf beetle. The reactions in these lines involves the tolerance and non-preference types of resistance.

Approximately 500 wheat varieties selected for cereal leaf beetle resistance under field nursery conditions were tested for resistance in the laboratory, utilizing growth response, survival of first instar larvae, ovipositional preference, and egg production of mated females as measures of resistance. Eighteen spring wheat varieties were found with high levels of resistance to both the larval and adult forms of the beetle. Plant pubescence was coincidental with the highly resistant wheat varieties. Laboratory and field results on cereal leaf beetle resistance have been complementary in most instances. Winter wheat CI 8519 and the 18 spring wheat varieties rated as the top lines in the lab tests were the best lines in the field nursery.

In recent tests conducted at Stillwater, Okla., Dickinson Selection 28A wheat appears to have lost its resistance to the greenbug. Previously it was susceptible only to a biotype that occurred only under greenhouse culture conditions. During the past year DS 28A was susceptible to Stillwater greenhouse greenbug cultures and field-collected cultures from Wichita and Manhattan, Kans. There was no difference in rate of reproduction on resistant DS 28A and susceptible Ponca wheats, whereas, the rate was reduced 45% on resistant Will barley as compared to susceptible Rogers barley. It is not known whether this loss of resistance is due to changes in the plant or in the greenbugs. Further tests are being made with greenbug cultures from Texas.

At Tifton, Ga., 36 sorghum lines were screened in the greenhouse for leaf damage by fall armyworm larvae. Line SA-392 (a Hegari type) exhibited the least damage (most resistance), while line FC-8962 (Kafir type) was the next most resistant.

Studies on physiological plant responses in relation to greenbug feeding on resistant barley plants were conducted at Brookings, S. D. Measurements were made of rate of photosynthesis, rate of respiration, chlorophyll content, fresh weight, dry weight, and area in relation to the amount of insect feeding. Comparisons of uninfested and infested leaves showed that chlorophyll content and rate of photosynthesis (based on fresh and dry weight and leaf area) decreased markedly in relation to increases in amount of insect feeding. Rate of respiration of infested leaves appeared slightly higher than uninfested leaves. Relationships between dry weight, fresh weight, and area of infested as compared with uninfested appeared to be unaltered during the course of this study.

Approximately 37 new crosses to introduce resistance to cereal leaf beetle into 6 commercial hard red spring wheats have been made at Brookings, S. D. A total of 580 F₃ lines from cereal leaf beetle resistant line x commercial spring wheat are available for testing in Michigan in spring of 1966. Four to five hundred lines from cereal leaf beetle resistant lines x commercial hard red winter wheats will be available for fall seeding.

Studies conducted under P.L. 480 project A7-ENT-31, in India, on investigations on the insect pests of sorghum and millets, show that the extent of loss due to shot fly, Atherigona indica, and stem borer, C. zonellus, was as high as 77.6% of grain. Two thousand and twenty-eight varieties of bajra were screened for resistance to insect pests. Some of these appear to be of good sources of insect resistant germ plasm for use in the breeding program.

H. Insect Vectors of Disease

1. Corn Insects. Dalbulus maidis, a vector of corn stunt virus, was collected in Texas on January 8, 1965, and in Louisiana in mid-July. This leafhopper had migrated eastward across Mississippi, where large field populations developed late in the season, and into Alabama by August 31, 1965. The leafhoppers were killed by below freezing temperatures in December. As of June 27, 1966, D. maidis has not been collected.

In tests conducted at State College, Miss., 3 native species of leafhoppers, Graminella nigrifrons, Deltocephalus flavicostus, and Exitianus exitosus, are being studied as possible vectors of corn stunt virus. All 3 are apparent vectors, producing virus-like symptoms in plants on which they are allowed to feed. D. maidis, a proven vector of corn stunt disease, completed several life cycles on gamagrass, Tripsacum dactyloides, a new host plant for this species.

In Mississippi, 669 entries composed of inbred lines, single and double crosses, commercial hybrids, and exotics, were tested for resistance to corn stunt virus. Different degrees of resistance were observed in native or locally adapted material indicating that unadapted resistant exotic material will not have to be used in breeding programs to develop resistant materials.

Field observations have indicated that the use of resistant hybrids by farmers in affected areas in 1965 has prevented the spread of corn stunt virus. A tabulation of commercial hybrids showing resistance to corn stunt virus was compiled for release to farmers.

In studies conducted under P.L. 480 project A10-ENT-5, Israel, on the hostplant-vector and hostplant-virus relationships in the rough dwarf virus disease of maize, it was discovered that D. striatella was the insect vector that carries and spreads the virus in the field. Subsequently, two more species of the same family were incriminated as vectors. These are D. pellucida and Sogatella vibix. The former species has a worldwide distribution and occurs also in North America, whereas the latter is apparently endemic in the Eastern Mediterranean, as it has hitherto been found only in Israel.

Studies were conducted at the University of Missouri under a grant on leafhopper vectors of corn viruses. A survey of insect vectors has been completed. Laboratory studies were conducted to develop techniques for rearing leafhoppers for transmission studies.

2. Small Grain Insects. A Delphacodes spp. collected at Brookings, S. Dak., late in the fall has been successfully colonized on rye. General biological studies including life history and host range investigations are being made of this insect. Such information would be of extreme value in the event that maize rough dwarf virus (MRDV) should be inadvertently introduced into the United States. American hybrid corns are known to be most susceptible in areas where MRDV occurs. This virus is only known to be transmitted in nature by species of Delphacodes.

Barley yellow dwarf virus has been successfully transmitted to and recovered from 5 varieties of sweet corn using the bird-cherry oat aphid, R. padi as the vector and a field isolate of BYDV collected at Davis, S. Dak. In all cases, the virus was recovered from test plants with non-infective aphids and serially transferred back to Black Hullless barley plants. Corn varieties were Aunt Mary, Golden Midget, Golden Sunshine, Golden Charlevoix, and Rainbow. This is the first report of transmission of BYDV to corn in the U. S. where infection was definitely established by serial recovery of the virus.

Rearing techniques for the leafhoppers, G. nigrifrons and D. sonorus have been worked out enabling the life histories to be studied in the greenhouse. G. nigrifrons completes its life cycle in an average of 34 days with approximately 13 days in the egg stage and about 4 to 6 days in each of the 5 nymphal instars. The host range study indicates that the majority of the cultivated small grains, corns, and sorghums are acceptable as both food and breeding hosts.

Field-collected aphids were pooled in groups of 25 aphids and screened for barley yellow dwarf disease throughout the autumn. A total of 35 pools of R. padi - R. rufiabdominalis, 8 pools of Forda olivacea, and 6 pools of M. avenae were screened. One pool of R. padi collected in late October transmitted BYD.

Barley yellow dwarf virus was recovered from migrating corn leaf aphids at Brookings, S. Dak. Major flights of corn leaf aphid, R. maidis, occurred during the first week of August. Five pools of 50 live aphids were formed from Johnson-Taylor trap catches and placed on healthy barley plants. Plants supporting two pools showed symptoms of barley yellow dwarf disease.

One hundred and fifty-two varieties of wheat were evaluated for barley yellow dwarf resistance in greenhouse tests in South Dakota. Ten wheat entries showed resistance to barley yellow dwarf. Fourteen of more than 1800 wheats in a field trial, showed good resistance to BYDV in field tests. These varieties have been incorporated into the winter and spring varieties breeding programs to develop commercial wheats with BYD resistance.

Twenty-nine hard red spring wheat varieties infested with BYVD in field plots at Brookings showed an average reduction in yield of 35.3%. Number of kernels per head and weight of kernels accounted for most of the reduction. The number of heads per foot of row appeared to be little affected by the virus in this trial.

Studies conducted under P.L. 480 project E8-ENT-1, Finland, indicates that Calligypona sordidula and C. obscurella were vectors of OSDV, oat streak dwarf virus, and WSMV, wheat streak mosaic virus, and Dicranotropis hamata the vector of OSDV. The injury caused to oats by C. sordidula was proved to be a virus disease. It was determined that the primary factor tending to increase the population of C. pellucida in its optimum area of occurrence is the constant cultivation of only cereals and hay. The most important factors reducing the population are natural enemies, winter mortality, and drought.

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RICE INSECTS
Entomology Research Division, ARS

Problem. Several species of insects including leafhoppers, the rice stink bug, rice water weevil, grape colaspis, stalk borers, and the sugarcane beetle damage rice in the rice-growing areas of the United States. Progress has been made toward the solution of some of the insect problems encountered in the production of rice but more effective, more economical, and safer insect control measures are needed. The appearance of resistance to certain insecticides in some rice insects stresses the need for basic information to overcome this problem. Additional emphasis should be given to new approaches to control rice insects and to evaluate rice varieties for resistance to major rice insects.

USDA AND COOPERATIVE PROGRAM

The Department's program on rice insects involves entomologists, agronomists, and plant breeders, and plant pathologists engaged in both basic studies and in the application of known principles to the solution of growers' problems. The research is being conducted at Baton Rouge, La., in cooperation with the Louisiana Experiment Station. Basic research on the control of damage by larvae of the rice water weevil, Lissorhoptrus oryzophilus, by increasing plant tolerance, is being conducted under a grant with the University of Arkansas, Fayetteville, Ark.; under PL 480, A7-ENT-5, India, a survey is being made of the natural enemies of pests of paddy.

The Federal scientific effort devoted to research in this area totals 1.2 scientists man-years. Of this number 0.3 is devoted to basic biology of the leafhoppers, rice stink bug, and rice water weevil; 0.3 to insecticidal control of rice stink bug and rice water weevil; 0.3 to varietal evaluation of rice for resistance to stink bug, rice water weevil, and vectors of rice diseases; 0.1 to insect vectors of hoja blanca; and 0.2 to program leadership. In addition Federal support for research in this area provides for 0.4 man-year in a research grant to the University of Arkansas for varietal evaluation of rice for resistance to the rice water weevil.

PROGRAM FOR THE STATE EXPERIMENT STATIONS

A total of 3.4 professional man-years is devoted to this area of research.

PROGRESS -- USDA AND COOPERATIVE PROGRAM

A. Basic Biology, Ecology and Nutrition

At Baton Rouge, La., a technique was developed for rearing the rice water weevil, Lissorhoptrus oryzophilus, in the greenhouse. This is the first time the weevil has been cultured under artificial conditions. Oviposition studies indicated that the rice water weevil deposited its eggs both below

and above the water line, predominantly in the first or second leaf sheath of the young rice plant. The maximum number of eggs per plant occurring 2 or 3 weeks after flooding. A field study on rice planted at weekly intervals over a period of 5 weeks, beginning April 20 showed that over 4 times the number of eggs were deposited in older seedlings (April 20 planting) than in the younger seedlings (May 18 planting) when egg counts were made 2 weeks after flooding.

Clumps of bullgrass, broom sedge, and smut grass collected in September 1965 at Crowley, La., averaged 4.7 diapausing rice water weevils per clump. There was no significant difference in the number of weevils on the 3 grass species.

Observations on overwinter larvae of Chilo plejadellus and Diatraea saccharalis in rice stubble, during 1965-66, showed that live D. saccharalis were rarely found in March and the overwintering populations of C. plejadellus were reduced about two-thirds. The tendency of D. saccharalis to overwinter at the bottom of the stalk where it is subjected to severe weather conditions probably accounts for high winter mortality in this species.

In studies conducted in India under PL 480 project A7-ENT-5, 36 species of insects were recorded as pests of paddy. Of these, 9 species of Lepidopterous borers were responsible for major losses to the paddy borer. Tryporyza incertulae was the most injurious and occurred throughout the rice-growing areas. The other borers in order of their importance were: Sesamia sp., Scirpophaga sp., Chilo partellus, Pseudaletia separata, Ancylolomia indica, Spodoptera cilium, Brachmia arotraea, and Leucania loreyi.

B. Insecticidal and Cultural Control

Nine experimental insecticides were evaluated for systemic action against Sogata orizicola. All materials except Niagara NIA-10242 exhibited some degree of phytotoxicity when used as seed treatments. Bay 39007 gave excellent control for 25 days following planting; however, both this material and Niagara NIA-10242 were incompatible with propanil, a commonly used herbicide in rice. Di-Syston, Isolan, and Phorate applied as a granular formulation in soil treatments at 2 pounds of toxicant per acre gave good control up to 33 days after treatment--No material was adequate at the one pound toxicant per acre rate.

Bidrin, carbaryl, phosphamidon, and Azodrin applied as a spray at the rate of one pound actual per acre gave good control. Solutions of 46 ppm of Bidrin, dimethoate, and Isolan, simulating the addition of insecticides to the flush water in a rice field, gave 50% control up to 15 days after treatment.

Granular formulations of Bay 25141 and Niagara NIA-10242 applied prior to flood at rates of 2 and 0.6 pounds per acre, respectively, gave adequate control of rice water weevil larvae. One application of as little as 0.3 pounds per acre of Niagara NIA-10242 also controlled the weevil. Niagara NIA-10242 and Temik showed sufficient systemic activity to cause significant reductions of the green rice leafhopper, Draeculacephala portola 6 days after flooding.

Investigations on the resistance of rice water weevil populations to aldrin revealed resistance occurring only on the Rice Experiment Station, Crowley, La., and at Jones and Alsatia in the northern part of Louisiana.

In germination tests, 5 insecticides had no effect on seed viability. All except AC 52160 and Bay 44646, lowered the viability of the seed, when seeded in water.

Seven insecticides and one combination of two insecticides were evaluated as seed treatments in the field against rice water weevil at both Crowley, La., and Stoneville, Miss. Aldrin at 0.25 pound (the recommended rates) and one pound per cwt. failed to control rice water weevil. The combination (aldrin-heptachlor) treatment was also unsatisfactory. Leafhoppers, D. portola, Graminella nigrifrons, and several other species were controlled with Niagara NIA-10242 seed treatments.

A technique for surveying for populations of the rice water weevil was also developed. Submersion of grass clumps containing weevils activated the diapausing adults which could then be readily recovered.

C. Biological Control

In studies conducted in India under PL 480 project A7-ENT-5, 33 species of parasites were recorded from paddy pests. Rhaconotus oryzae, Tropobracon schoenobii, Bracon chinensis, and Chelonus sp. were the commonest and fairly widely distributed parasites of the borers. Telenomus sp., R. oryzae, Goniozus sp., Glyptomorpha deesae, Isotima sp., and Hockeria sp. appeared to be new records on paddy borers in West Pakistan. Parasitism was higher on the larvae in stubbles than on those in the crop. Although a parasitism of up to 27.7% of the larvae of T. incertulas on 'Sathra' crop in May in Zone 3 and up to 41.1% of those in stubbles of 'Kangni' 27 in June in Zone 4 was recorded, the average parasitism on borers in all the ecological Zones as a whole was very low.

D. Varietal Evaluation for Insect Resistance

Seventy varieties or crosses representing either the commercial varieties in production or the more advanced potential varieties available from breeders of the southern rice belt were evaluated for resistance to both rice water weevil and borers. Marked differences between selections were apparent relative to rice water weevil larval infestation. Colusa and some

of the crosses with Rexora parentage received low infestation. Nova, Saturn, Zenith, Nato, Gulfrose, Century Patna 231, Fortuna, Bluebonnet 50, Toro, Arkrose, Lacrosse, Vegold, and the newly released variety, Dawn, had intermediate infestations. Early Prolific, Blue Rose, Supreme Blue Rose, Improved Blue Rose, and Calrose had heavy infestations. Crosses of PI215936 X C19214 from both the early and the midseason maturing long and long-slender grain rice variety types had low infestations. No significant differences were found in the resistance of these varieties/crosses to Chilo and Diatraea.

Preliminary results of field tests conducted under a grant by the University of Arkansas, indicated that higher nitrogen levels are associated with high rice water weevil larval infestations. Greenhouse tests indicated that the adult weevils prefer to feed upon the younger leaf blades which contain more sucrose and starch and less cellulose than older leaves. The weevils preferred older leaves for oviposition. The leaf-sheath was the preferred site of oviposition.

In India under PL 480 project A7-ENT-5, studies on 20 varieties of paddy including 8 fine and 12 coarse (16 native and 4 from the Philippines) have been carried out. Only one variety, 'Sonkari Kangni' (native), remained free from borer attack, while 'Daudzai' (native) had the highest average infestation during the season. Among the fine varieties 'Dokri Basmati' had the least borer attack, while 'Basmati' C621 and C622 had infestations of medium intensity. The other fine varieties were fairly heavily infested. On the whole late maturing varieties were more heavily attacked as compared with early ones.

E. Insect Vectors of Diseases

Progeny, of reciprocal matings of HBV transmitters to non-transmitters, were reared from the egg under aseptic conditions and constant temperature and photoperiod. Planthoppers that were progeny of non-transmitting females, transmitted HBV, although they had not fed on HBV diseased plant sources indicating masked virus infectivity. Adult planthoppers that acquired virus transovarially died sooner than others.

Two thousand and ninety-seven males and 863 females, active vectors of hoja blanca virus, were reared for use in cooperative tests with Crops Research Division to develop varieties resistant to this disease.

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FORAGE AND RANGE INSECTS
Entomology Research Division, ARS

Problem. Numerous insect pests that attack forage and range plants in various parts of the United States lower seed production, reduce the quantity and quality of forage crops, and decrease the abundance of range plants for the grazing of livestock. Certain insects are involved in the transmission of forage-crop diseases. Among the more important insect pests are grasshoppers, lygus and other plant bugs, stink bugs, seed chalcids, the alfalfa weevil, root borers, spittlebugs, leafhoppers, and a variety of aphids including the spotted alfalfa aphid and the pea aphid. A variety of insecticides is used to control these insects but they are often costly and may create residue hazards in meat and milk as well as adversely affect wildlife. There is great need for more efficient insecticides that can be applied on forage crops and range vegetation without leaving residues harmful to man or animals or that might harm bees and other pollinating insects. Increased attention should be given to the development of non-chemical control methods. The search for insect parasites, predators, and pathogens and ways to employ them effectively should be emphasized in research. The development of crop varieties which resist attack by insects offers economical and safe insect-control procedures. Forage crops should be evaluated for resistance to major insect pests and resistant germ plasm should be made available for use by the plant breeders in crop-improvement programs. Basic studies are also needed on the feeding habits of grasshoppers under different environments that affect the abundance of these insect pests. New approaches to control of forage and range insects, such as sterilization techniques and sex attractants, should be investigated.

USDA AND COOPERATIVE PROGRAM

The Department has a continuing long-term program of basic and applied research on forage and range insects. Studies on varieties of alfalfa resistant to insects are cooperative with State and Federal agronomists and plant breeders, those on plant disease transmission by insects with plant pathologists, and research on insecticide residues with chemists. Grasshopper research at Bozeman, Mont., and Mesa, Ariz., is cooperative with the respective State Experiment Stations. White-fringed beetle research is conducted at Gulfport, Miss. Biological control studies on armyworms and cutworms at Baton Rouge are cooperative with the Louisiana Experiment Station. Investigations on alfalfa insects are being conducted at Mesa, and Tucson, Ariz., Lincoln, Nebr., and Beltsville, Md., in cooperation with the Experiment Stations in these States. Research on clover and turf grass insects at Forest Grove, Oreg., is conducted in cooperation with the Oregon Experiment Station. Work on grass insects, plant disease transmission by insects, and insecticide residues at Tifton, Ga., is cooperative with the Georgia Experiment Station. Research on insecticide residues is conducted at Beltsville, Md., and at Yakima, Wash.,

in cooperation with the Washington Experiment Station. Studies on varietal resistance, insect vectors of plant diseases, and grass insects at University Park, Pa., is cooperative with Experiment Stations in 12 Northeastern States. Certain phases of the research on forage and range insects are contributing to regional projects W-37 (Natural Factors Responsible for Grasshopper Population Changes), NC-52 (Factors Influencing the Distribution and Abundance of Grasshoppers), W-74 (Seed Chalcids Attacking Small-Seeded Leguminous Crops), and S-55 (Alfalfa Insects).

Two contracts, 10 grants, and 1 cooperative agreement with State Universities and Experiment Stations will provide additional research on insect biology, physiology, and nutrition, biological control, attractants, and varietal resistance.

The Federal scientific effort devoted to research in this area totals 24.0 scientist man-years. Of this number 4.5 man-years are devoted to basic biology, physiology, and nutrition, 5.0 to insecticidal and cultural control, 4.0 to insecticide residue determinations, 4.0 to biological control, 0.6 to insect sterility, attractants, and other new approaches to control, 0.5 to evaluation of equipment for insect detection and control, 4.1 to varietal evaluation for insect resistance, 0.2 to insect vectors of diseases, and 1.1 to program leadership.

In addition Federal support of research in this area under contracts and grants provides a total of 3.2 scientist man-years. Of this total 0.9 is devoted to basic biology, physiology, and nutrition, 0.6 to biological control, 0.3 to insect attractants and other new methods of control, and 1.4 to varietal evaluation of insect resistance.

Two P.L. 480 projects are underway: E21-ENT-9, "Insect Vectors of Virus Diseases of Various Forage Legumes" with the Research Institute of Plant Protection, Poznan, Poland, and A10-ENT-6, "Acoustic Responses of the Desert Locust (*Schistocerca gregaria* Forsk.), Moroccan Locust (*Dociostarus maroccanus* Thbg.), and *Acrotylus insubricus* Scop. (Orthoptera, Acrididae)" with the Hebrew University of Jerusalem, Jerusalem, Israel.

PROGRAM OF STATE EXPERIMENT STATIONS

A total of 44.5 professional man-years is devoted to this area of research.

PROGRESS -- USDA AND COOPERATIVE PROGRAM

A. Basic Biology, Physiology, and Nutrition

1. Grasshoppers. Severe drought and poor vegetation conditions in the spring of 1964 and the summer of 1965, reduced the desert grasshopper population in Arizona, but above-normal rainfall and better condition of desert vegetation in late 1965 and the early part of 1966 was favorable for an increase in this grasshopper species in 1966.

The average grasshopper population in rangeland habitats at Peeples Valley, Ariz., increased 73% in 1965 over that in 1964. This was the second successive increase. Winter and spring rainfall was considerably above normal and vegetative ground cover increased from 69% in 1964 to 77% in 1965.

In 1965 in Arizona, the grasshopper population increase in Conservation Reserve land was 368% over 1964, and in weedy field margins the increase was 330%.

To determine the affect of crowding on the migratory grasshopper, groups were reared in cages at the rate of one to 3.5 cu. in. and at one to 0.6 cu. in. There was no significant difference in the average length of the nymphal period, percentage survival, and body measurements and front wing deformity of resulting adults, at the 2 nymphal densities.

The percentage of migratory grasshopper eggs that hatched was greater for females that were exposed to males of the same age throughout the adult period than for females that were exposed to males for only the first 29 days of adult life, and still greater than for females that were exposed to males for only the first 17 days of life. The shortest adult longevity was in the group of females exposed to males throughout the adult period. Hatching percentages were low for eggs laid during the latter part of the oviposition period regardless of the length of the period the adult females were exposed to males.

Range grasshopper studies were continued in 1966 on several shortgrass rangelands in central and southeastern Arizona to determine the effects of weather, type, and condition of vegetation, range usage and management practices, and natural enemies on seasonal history, abundance, and plant damage of important grasshoppers. Populations of spring-group and summer-group grasshoppers were low owing to dry plant conditions in June and early July 1965 that were unfavorable for grasshopper development and survival.

Studies were continued on two 20-acre range recovery plots. Each plot is subdivided into 2 fenced and 2 unfenced 5-acre plots. On a sparse grass area for the period April-July 1965 average square-yard grasshopper densities in untreated fenced and grazed plots were 1.51 and 1.44, respectively. Loss of weedy and grass herbage in spring was negligible due to an abundance of vegetation; whereas on a comparable date in 1964 the loss of weedy forage was about 95%.

Research was initiated in Israel under PL 480 project to study the acoustic response of grasshoppers. Sounds emitted by male grasshoppers are made of compound pulses; the number of these in 0.5 seconds is called pulse rate. There was a marked increase in the pulse rate within the temperature range of 20° to 35° C whereas there was no marked increase at the 40° and 45° C. The age at which males started to emit stridulatory sounds was directly related to the temperature at which they

were reared. At 25° C they emitted sounds 24 days after reaching the adult stage, at 30° C 16 days, and at 44° C 5 days.

A grant was recently awarded Kansas State University to determine the food habits of selected grasshopper species when allowed a choice between range-inhabiting plants and cultivated pasture plants.

2. Alfalfa Insects. In preliminary studies at Lincoln, Nebr., to determine if sexual forms of the spotted alfalfa aphid could be produced in environmental chambers, the test insects were subjected to day and night temperatures of 70 and 60° F., respectively, and 8-1/2 hours of light. After 8 generations under these conditions no egg laying forms appeared. Prior field observations have indicated that there are 2 strains of the spotted alfalfa aphid; one with the ability to produce sexuales when subjected to the proper conditions and the other without this ability.

At Tucson, Ariz., efforts were made to determine conditions under which a year-around supply of larvae of the Egyptian alfalfa weevil could be achieved for use in plant resistance studies. It seemed probable that the small percentage of non-diapausing F₁ adults obtained by rearing weevils under a short photoperiod was partially the result of rearing them at high temperature (90° to 70° F). The culture was lost when only 4 F₂ adults were reared and all diapaused. These studies indicate that diapause is obligatory in most individuals or that factors other than temperature and photoperiod are involved.

At Beltsville, Md., the non-diapausing laboratory colony of the alfalfa weevil started in 1961 has completed its 23rd generation with no apparent loss of vigor. The larvae are reared under 8 hours of light and 16 hours of darkness to prevent diapause.

The suitability of 7 legumes as hosts for the alfalfa weevil was determined in the laboratory. All have been reported as being naturally infested under field conditions. The criteria used were adult feeding, survival from egg to adult, and oviposition, all in isolation. Significant host differences were found for all 3 criteria with alfalfa and sweetclover being most suitable and equally so. All 3 factors were found to be significantly and positively correlated. Alsike clover, ladino clover, red clover, and hairy vetch were relatively unsuitable as hosts. The presence of a dietary oviposition stimulus in alfalfa and sweetclover was definitely demonstrated.

The attractiveness of steam-distilled volatile components of these legumes plus hop clover was determined in a series of concentrations. Only 3 (alfalfa, alsike clover, and red clover) produced significant attractancy. Sweetclover and hairy vetch were unattractive. Ladino clover and hop clover were repellent.

Periodic surveys in 10 alfalfa fields for adult weevils showed a 70% decline from the winter of 1964-65 to the winter of 1965-66. Average populations per square foot were 9.4 and 3.9 for November and March of 1964-65, and 2.9 and 1.1 for the same months of 1965-66. Larval populations tended to decline also with peaks in individual fields, 1000 per square foot in the spring of 1965, but only 580 in the spring of 1966. This is the first indication of a population decline in the East since the weevil was discovered in Maryland in 1952. Adult weevils apparently return to alfalfa fields in the fall at random. There is no correlation between numbers of adults per square foot and age of alfalfa stand, amount of fall growth, stand density, or surrounding habitat.

A grant was recently awarded to New Mexico State University to develop an artificial rearing method for sucking insects and to determine their nutritive requirements. Work to date has been to develop techniques for feeding aphids and lygus bugs through an artificial membrane.

A grant was recently awarded to the University of California to determine the toxic ingredient in the salivary glands of lygus bugs that cause injury when these insects feed on alfalfa.

3. Clover Insects. Experiments were conducted at Lincoln, Nebr., to determine if sexuales of the sweetclover aphid could be produced in the laboratory. Sexual forms containing eggs were produced after 2 to 3 generations at photoperiods of 8-1/2 hours and day-night temperatures of 70 and 60° F.

4. Grass Insects. A brome-grass seed midge, which has been under study at Lincoln, Nebr., since 1965 has now been identified as Stenodiplosis bromicola. This species, which is recorded as a serious pest of brome-grass seed in Russia, had not previously been known to occur in the United States. This species appears to be widespread within Nebraska--adult midges were collected in 4 east and central counties and damaged seed heads were collected in 11 counties from east to west across the southern part of the State. The insect appeared to enter diapause as the brome-grass seed reached maturity during the 1965 season. Adults were again present and active in eastern Nebraska brome-grass fields in 1966.

Studies showed that adult two lined-spittlebugs less than 1 day old caused less damage to Coastal Bermudagrass than those 1 to 5 days old. There was no difference in the amount of damage caused by unmated females and males 1 to 25 days old. However, 7 day old mated females caused more damage than 7 day old unmated females. A large proportion of female two-lined spittlebugs collected from light traps contained no eggs in their ovaries, suggesting that older females with mature ovaries probably do not fly to traps.

At Tifton, Ga., the fall armyworm was reared on millet grown in soil with varying fertility levels. In general, larvae fed millet from high nitrogen levels were heavier at 6 and 9 days of age than those fed millet

from other fertilizer levels. Pupal weights did not follow the same order as larval weights and the differences were less significant. The larval period to pupation was about 3 days longer for larvae fed millet grown on unfertilized soil than those fed millet grown on high fertilization levels.

At Tifton, Ga., pearl millet breeding experiments indicate that pollination is accomplished not only by wind, but probably also by insects. The major insect visitors in the vicinity of Tifton in 1965, were the honey bee, a bumble bee, Bombus impatiens, and a cantharid beetle, Chauliognathus marginatus.

At Forest Grove, Oreg., 11 species of sod webworms were found infesting lawn and turf areas. However, all of the major damage to commercial grass plantings in Oregon has been caused by one species, Crambus toparius. This species is present from late May to early August and reaches peak abundance in mid-June. Several other species were present in low numbers in commercial plantings. Of the species infesting lawn areas, C. bonifatellus was most abundant and was present throughout the season. Damage by this species was difficult to find in lawns possibly because regular irrigation and fertilization of well kept lawns enabled the grass to outgrow the damage. A few species were found infesting range areas but in non-economic numbers.

5. White-fringed Beetles. At Gulfport, Miss., a laboratory technique has been developed for testing feeding preferences of white-fringed beetles when the adults are offered a choice of several species of plants.

All legumes tested were fed upon heavily. Other preferred hosts were rose, althea, gardenia, ivy-leafed morning glory, pecan, dahlia, cotton, and teaweed. Such plants as bamboo, bitterweed, broomsedge, dog fennel, fern, sow thistle, bull thistle, and yucca were not fed upon.

An artificial diet which appears to be equal to natural diets for white-fringed beetle adults has been devised in Alabama under a research grant with Auburn University. All artificial diets developed for white-fringed beetle larvae have been unsuccessful primarily because mold inhibitors required to prevent mold growth on the diet were lethal to the larvae.

B. Insecticidal and Cultural Control

1. Grasshoppers. Tests of low-volume applications of 6 insecticide formulations were made by airplane for grasshopper control of rangeland in Montana. Total volume per acre ranged from 7 to 21.3 ounces. Technical malathion at 6 ounces per acre was used as a check. Although all insecticides gave satisfactory control under some conditions, meteorological factors and physical characteristics of the spray materials were found to be of much greater significance in the low-volume method of treatment than with previously used larger volumes. For example the very fine

atomization of diazinon reduced its control, whereas fine atomization of naled appeared desirable.

At Bozeman, Mont., 5 insecticide formulations applied by low-volume ground equipment were compared with malathion for grasshopper control. Two were approximately equal to the malathion standard and 3 gave better control.

Laboratory screening tests at Bozeman, Mont., were conducted to determine the effectiveness of 34 candidate insecticides against adults of the migratory grasshopper. By use of a dosage-mortality regression line for analytical grade malathion, the approximate 80-90% dosage levels were determined for males (3.90 ug/insect) and females (5.80 ug/insect). These dosages were used for the candidate insecticides and the malathion standard. Acetone solutions of the insecticides were applied topically, or to lettuce discs in cases where oral tests were conducted, by means of a calibrated micro-syringe. Following treatment the grasshoppers were confined in screen cages with lettuce as food. Mortalities were recorded after 24, 48, and 72 hours. Three candidate insecticides were better than malathion and 2 were equally as effective.

2. Alfalfa Insects. Small plot tests were conducted at Beltsville, Md., comparing 28 experimental insecticide treatments with a methoxychlor-malathion standard to control the alfalfa weevil. All were applied to the growing crop in May 1966. Eight materials showed 5% or less foliage damage 20 days after application and seven other materials showing less than 10%. The addition of parathion to Shell SD-7438 and Sevin did not increase control, nor did the addition of malathion to methoxychlor.

Seven materials were compared for alfalfa weevil control on large plots. All were equally effective 7 days after application. Larval counts 20 days after application showed only azinphosmethyl (3/4 lb), Imidan^R (1 lb), and methoxychlor (1-1/2 lb) significantly better than the untreated check. Other materials applied were carbaryl, malathion, parathion, and diazinon in order of their effectiveness.

At Lincoln, Nebr., 7 insecticides were initially equally effective in reducing nymphs of the tarnished plant bug, but DDT was effective over a longer period. None of the treatments provided control for the alfalfa seed chalcid. DDT increased the yield of seed by 163% over that of the check. None of the other treatments significantly increased yields.

In tests in Arizona to control the red harvester ant in alfalfa fields, Kepone bait gave 80% control of ant colonies 3 months after application compared with only 45% control with mirex bait.

3. Clover Insects. Since dieldrin has been withdrawn for use on sweet-clover, tests have been initiated at Lincoln, Nebr., to find other insecticides for control of the sweetclover weevil. Bay 39007 is the only one

of 5 insecticides tested that has shown promise. Additional tests will be necessary to determine if it will protect the seedlings long enough to be of practical value.

4. Grass Insects. At Tifton, Ga., Coastal bermudagrass plots that were mowed regularly for hay had little to no spittlebug damage while plots that were not mowed regularly had high numbers of spittlebugs and were badly damaged.

At Tifton, Ga., one of two granular insecticides applied at the rate of 1 pound per acre in each of 2 applications to 2 separate fields of Coastal bermudagrass in August 1965 to control an insect complex increased the yield 1/3 ton dry weight more per acre as compared to the check.

At Tifton, Ga., 4 of 6 insecticides applied to Coastal bermudagrass at 1 pound per acre gave good control of Mocis spp.

At Tifton, Ga., 7 insecticides applied at 2 pounds per acre were evaluated for control of mole crickets in millet. Effectiveness was based on dead crickets, tunneling, stand reduction, and yield. The 4 best insecticides, based upon the various indices of effectiveness were trichlorfon granules, Mobil MC-A-600 granules, Stauffer N-2790 bait, and Kepone bait.

5. White-fringed beetles. In an effort to develop an insecticide screening method for white-fringed beetles, newly hatched larvae were placed in direct contact with heptachlor, dieldrin, or DDT and older larvae were dipped in acetone solutions of several insecticides. The younger larval were not killed and results with the older larvae were inconsistent.

Balled and burlapped nursery plants (ornamentals) infested with white-fringed beetle larvae were dipped in solutions of ethylene dibromide formula No. 440 and ethylene dibromide chlordane formula No. 431 for about 15 minutes. Two weeks later all larvae were dead in plant balls treated at the 4 ml/gal dosage of each. A few larvae were classified as moribund in plant balls treated at the 2 ml/gal dosage. No apparent phytotoxic effect was noted in any of the plants.

C. Insecticide Residue Determinations

1. Mobil MC-A-600. Samples of milk from cows that had been fed Coastal bermudagrass silage which had been sprayed with Mobil MC-A-600 at levels up to 32 ounces per acre were analyzed. No detectable residues were found. The concentration of Mobil MC-A-600 in bermudagrass silage that had been treated at the 32-ounce-per-acre dosage decreased from 63 ppm on the date the grass was ensiled to 34 ppm after 36 days in the silo.

2. Bomyl[®]. Analyses were made of samples of millet, corn, and Coastal bermudagrass from plots at Tifton, Ga., treated with emulsion sprays of Bomyl[®]. Millet treated with 4 ounces per acre contained residues of 0.63 ppm the day it was sprayed and less than 0.1 ppm one day after treatment. Millet treated with 8 ounces per acre contained residues of 0.16 ppm one day after treatment. Corn samples collected 2 days after treatment contained less than 0.1 ppm. Residues on Coastal bermudagrass from both levels of treatment were in excess of 1 ppm one day after treatment, but were less than 0.1 ppm 6 days after treatment.

3. Niagara NIA-10242. Residues of Niagara NIA-10242 on Coastal bermudagrass treated at the rate of 16 ounces per acre decreased from 43 ppm immediately after spraying to 2.1 ppm 14 days after spraying.

4. Malathion. At Bozeman, Mont., and Yakima, Wash., studies were made to compare residues of malathion applied to range grass at 2.5 ounces per acre as a solution in a light cracked-gas oil (1 gal per acre), a water solution (1 gal per acre), and a low-volume concentrate (1 pt per acre). The initial residues from the light cracked-gas oil solution, the water solution, and the low-volume concentrate, were 223, 177, and 160 ppm, respectively. Twenty-one days later these values had decreased to 34, 19, and 23.

D. Biological Control

1. Grasshoppers. In 1965, observations were continued in Arizona on natural enemies of grasshoppers. Parasitization by nemestrinid flies ranged up to 72% in some locations.

At Bozeman, Mont., efforts are being made to use microorganisms for control of grasshoppers. These include three protozoans and a virus.

Of the 4 pathogens, Nosema locustae (a sporozoan which infects the grasshopper fat bodies) has been used most extensively in laboratory and field studies. This organism was applied in the field under experimental conditions in 1964 and 1965. Although these applications have been successful in establishing the disease in the grasshopper populations, they have not been instrumental in causing detectable population reductions.

Another sporozoan, Nosema n. sp., has been isolated and described from grasshoppers. This organism infects the digestive tract, fat bodies, pericardium, gonads, and nervous system of the host insect. It completes its life cycle in 5 days, during which it induces the formation of tumors in the host. It is apparent that the relatively high pathogenicity of this organism is due to these tumors, which ultimately produce lesions in the digestive tract and cause the death of the host due to secondary bacterial septicemia.

Malamoeba locustae, an amoebic protozoan, infects the digestive tract epithelium and Malpighian tubules of grasshoppers. This results in decreased vitality and reduced fecundity in the host insect. The organism is a common problem in laboratory grasshopper cultures and studies are in progress to control the organism through the use of antibiotics. Triple sulfa has been effectively used for controlling the organism in infected specimens as well as preventing infections in young cultures.

During the past year a polyhedral virus was isolated from grasshoppers. This is the first instance of a virus being isolated from grasshoppers. This virus infects the fat bodies of grasshoppers which results in reduced fecundity and premature mortalities.

A grant was recently awarded to Montana State University to study the virus diseases that attack grasshoppers.

2. Alfalfa Weevil. In 1966, the alfalfa weevil continued to spread northward in Missouri, Illinois, and Ohio, and was found for the first time in Michigan. Parasitism of larvae by the 2 most widely established parasites, Bathyplectes curculionis and Tetrastichus incertus, during peak larval population in Maryland and New Jersey was low. Bathyplectes did not exceed 13% parasitism in any of the 20 survey fields until June when host populations were very low. Tetrastichus appeared commonly during June, as is normal. A bacterial disease of larvae was isolated in the fall of 1965. The isolate was highly pathogenic to larvae by injection, and slowly pathogenic when fed contaminated foliage. Larvae exhibiting the typical disease symptoms were collected commonly in the field throughout the spring of 1966.

A study, begun in the spring of 1963, to determine the amount and effect of parasitism on the alfalfa weevil, Hynera postica; in the area immediately east of the Rocky Mountains has been continued. Parasitism by B. curculionis in 1965 ranged from 31% in the Arkansas Valley (Kansas and Colorado) to 90% in the North Platte Valley (Nebraska and Wyoming). Weevil populations, although not economic, are increasing in the Arkansas Valley but are stable or decreasing in areas with higher rates of parasitism.

3. Egyptian Alfalfa Weevil. Collections of the Egyptian alfalfa weevil in areas where the larval parasite, T. incertus, was released in 1965, indicated that this parasite probably did not become established, and additional releases were made in 1966.

4. Spotted Alfalfa Aphid. Trioxys utilis was the only parasite of the spotted alfalfa aphid that was recovered in 1965 and 1966 in Arizona. No parasitized aphids were found in June but by August 2.4% were parasitized.

Systematic biweekly observations on the spotted alfalfa aphid and its natural enemies were continued until the end of 1965 in 4 alfalfa fields in the Salt River Valley of Arizona. In 1966, the leading predators of the

spotted alfalfa aphid in Arizona are Orius spp., Collops vittatus, and nabids. There was a 57% increase in total predators over 1964. Aphid mortality from fungus diseases was 18% early in December following a very rainy period, but it was not observed during the rest of the year.

5. Armyworms and Cutworms. At Baton Rouge, La., a study was undertaken to determine the possibility of using dissection for the determination of parasitism in two species of armyworms. Field collections of larvae of the armyworm and fall armyworm were divided into two lots. One lot was reared to determine parasitism, and the other lot dissected. Results with the fall armyworm were encouraging and percentages of parasitism by both methods were relatively close. In the case of Pseudaletia, however, nearly twice as many parasites were reared as were dissected.

Over a 5-year period in Louisiana, 17 species of armyworm parasites were identified. Although the overall percentage of parasites was very uniform from year to year (35 to 39%) the variation in the relative number of the various parasite species was great.

6. Rhodesgrass Scale. Studies were continued under a research contract with the Texas Experiment Station on mass producing and distributing Neodusmetia sangwani, a parasite of the Rhodesgrass scale. These tests have shown that as the number of female parasites was increased at each release point, the percentage of colony establishment increased. August and September was a better time for release of parasites than during the winter. For mass release of this parasite the method that appears most desirable is to collect grass from areas where the parasite is well established and place 10 to 15 stems in a paper box and make releases from an airplane at one box per square mile.

7. White-fringed Beetle. At Gulfport, Miss., microscopic examination of white-fringed beetle larvae that had been confined to dyed soil showed dye in the midgut and hindgut. This indicates that the larvae ingest some soil, and could ingest pathogens that might be present in the soil.

8. Grass Insects. At University Park, Pa., a Dryinid wasp was found parasitizing nymphs and adults of three species of leafhoppers and two species of plant hoppers collected from timothy and brome grass. Parasitism ranged from 1 to 25%. Parasitized hosts had from 1 to 3 Dryinid larvae. These left the host and spun a small grayish white cocoon on blades of grass and trash. All reared and collected adult Dryinid wasps were apterous females. Adult females are predacious.

E. Insect Sterility, Attractants, and Other New Approaches to Control

1. Spittlebugs. Although the female two-lined spittlebug gives off a pungent vanilla-like odor, workers at Tifton, Ga., determined that this odor was not a sex attractant, but acted more like a repellent.

2. Fall Armyworm. At Tifton, Ga., observations of the fall armyworm have revealed that crepuscular activity of the moths first involves congregation on host plants for feeding. After a period of feeding, the moths become quiescent for a time, then they often begin vibrating their wings before mating. Recordings on a sonagram of the wing vibrations of the roosting moths indicated the frequency was very low, in the 60-70 cps range. Sounds from magnetic speakers within this frequency range and directed into a cage of moths resulted in erratic results but a strong indication was obtained that moths were attracted to the sound.

3. Sweetclover Weevil. At Lincoln, Nebr., to investigate the attractiveness of coumarin to the sweetclover weevil, one square foot plots within an alfalfa field were treated by applying 200 mg of coumarin to the ground surface. Under the conditions of this test, coumarin was attractive to adult weevils as evidenced by their presence in the plots and by the fact that 15.4% of the alfalfa leaves in the treated areas had weevil feeding notches on them in comparison to 1.4% in equal areas of nearby untreated alfalfa.

4. Alfalfa weevil. In research conducted under a grant to Virginia Polytechnic Institute, 14 extracts from alfalfa were tested for attractiveness to the alfalfa weevil. A hot aqueous 2% NaCl extract was the most promising. Water extracts and ethanol extracts have also elicited some response.

5. Alfalfa Seed Chalcid. Studies conducted at the University of Wyoming under a research grant has shown that there may be chemosensory hairs on the alfalfa seed chalcid ovipositor which may influence ovipositional behavior. A total of 81 chemicals from the alfalfa plant have been tested and several have elicited an ovipositional response.

F. Evaluation of Equipment for Insect Detection and Control

1. Alfalfa Insects. Field research with liquid propane flammers was continued at Beltsville, Md., for control of the alfalfa weevil. Applications made in March have given control nearly equal to that obtained with a standard insecticide application of methoxychlor-malathion. Flaming stubble immediately after first harvest has also given good control without harm to the subsequent stand or yield.

G. Varietal Evaluation for Insect Resistance

1. Potato Leafhopper. In cooperation with the Crops Research Division, a technique developed at University Park, Pa., to screen alfalfa seedlings for resistance to meadow spittlebug, potato leafhopper, and alfalfa weevil, was used to screen two pools of alfalfa, each containing about 1130 plants. The best 90 plants were chosen from each pool and are being intercrossed.

At Lincoln, Nebr., a replicated alfalfa nursery of clones resistant, intermediate, and susceptible to the potato leafhopper was established in 1964

to study the relationship between leafhopper numbers, injury score (amount of yellowing), stage of growth, and changes in quality (protein, nitrate, carotene, dry matter, and fiber). One half of the nursery was treated weekly with insecticide in 1965. Protein content of both insecticide treated and untreated samples of the susceptible clone decreased with plant age. Total protein in the treated samples was, however, always as high or higher each time sampled. Protein content of the untreated intermediate clone increased with an increase of the injury score from 1 to 3 and then progressively decreased while the protein content of the treated samples of the same clone decreased evenly as plant age increased. Fiber content of untreated samples from both clones remained relatively constant with plant age. Carotene and nitrate content of the untreated and treated samples of both clones decreased with time. Carotene content of the treated samples was consistently higher than the untreated samples from both clones. Not much difference in the nitrate content was observed between the treated and untreated samples from both clones.

2. Alfalfa Weevil. Additional improvements have been made in laboratory testing techniques for screening for resistance to the alfalfa weevil at Beltsville, Md. Procedures and techniques are now fairly well standardized. Mass screening of seedling material in the cotyledon stage by exposure to adult feeding has proven effective in isolating promising material. Plants surviving this screening, and clonal material selected in the field, are then subjected in isolation to tests of (1) adult feeding on leaf disks, (2) larval survival and development, and (3) oviposition response. Significant differences have been found among entries for all three factors. Superior selections are being intercrossed in various ways in an attempt to intensify the level of resistance presently found.

3. Spotted Alfalfa Aphid and Pea Aphid. Testing of recently released commercial alfalfa varieties and promising experimentals composed by breeders of Arizona, California, and Nevada for resistance to biotypes ENT A and ENT B of the spotted alfalfa aphid shows that resistance is progressively improving in both public and private programs. Public varieties are still superior to private varieties in this regard but one private variety (Bonanza) ranked just behind the best public varieties.

In support of the alfalfa breeding programs of Arizona and California, 1300 seedlings highly resistant to the spotted alfalfa aphid were screened from populations of the experimental varieties SW-25, SW-39, and SW-40, all of which are of Sirsa origin. These seedlings are now being used to improve disease and insect resistance and agronomic quality of these varieties by reselection for other characters. Five hundred of these seedlings screened from SW-25 have been screened for pea aphid resistance at Tucson and 35 plants were selected for crossing and reselection among progeny.

Polycross seed of each of the 14 clones of experimental variety Nevada Syn X was screened for both spotted alfalfa aphid and pea aphid resistant plants. Ten progeny of each parent selected as highly resistant to both

aphids were returned to the breeder for a second polycrossing prior to rescreening.

Recent studies at Lincoln, Nebr., have shown that there is an interaction between four alfalfa clones and temperature in regard to resistance to the pea aphid and the spotted alfalfa aphid when the aphids were confined to the plants. Resistance breaks down as the temperature is lowered.

Eighty alfalfa plants selected in field nurseries for resistance to the potato leafhopper were tested for resistance to both the pea aphid and the spotted alfalfa aphid in the greenhouse. Of the 80, 18 were classified as resistant to both aphids or resistant to one and intermediate to the other.

Under a contract with Kansas State University good progress is being made in developing alfalfa varieties with resistance to two or more insects. Seed of one synthetic variety with resistance to pea aphid, spotted alfalfa aphid, and bacterial wilt is presently being increased in Oregon.

Work was continued under a long-term cooperative agreement jointly with the Crops Research Division and the Nevada Experiment Station to determine the biochemical nature of alfalfa resistance to the pea aphid and the spotted alfalfa aphid. The bioassay of extracted plant materials in artificial feeding devices has not been entirely successful, but it has been shown by P^{32} uptake in aphids that they feed more consistently on susceptible than resistant plants.

4. Alfalfa Seed Chalcid. At Mesa, Ariz., progeny tests on 14 plants resistant to the alfalfa seed chalcid showed that the level of resistance in the progeny was below that of the parents. This reduction in resistance was attributed to the introduction of foreign pollen from susceptible plants to the progeny during pollination.

5. Lygus Bugs. At Tucson, Ariz., no lygus bug resistant alfalfa varieties were found in a group of 116 tested.

At Manhattan, Kans., under a grant with Kansas State University, an attempt has been made to evaluate alfalfa in the seedling stage instead of in the blooming stage for resistance to the tarnish plant bug. Preliminary tests were inconclusive, but differences within and between varieties were obtained.

6. Sweetclover Aphid. Initially, results of studies at Lincoln, Nebr., on the inheritance of aphid resistance in sweetclover indicated only a single pair of genes. Additional data suggest the possibility of two pair of complementary genes governing aphid resistance.

7. Sweetclover Weevil. At Lincoln, Nebr., under a grant with the University of Nebraska, a bioassay method to help determine the factor causing resistance in Melilotus infesta to the sweetclover weevil has been

developed using sweetclover root disks processed by freeze drying. Water methanol extracts from M. infesta appear to cause a response from the weevil.

8. Grass Insects. At Tifton, Ga., 28 bermudagrasses were compared with Coastal bermudagrass for resistance to fall armyworm larvae. Three selections showed less feeding than the Coastal bermudagrass.

Emphasis is being given at Bozeman, Mont., to a study of grasshopper resistance in range grasses and legumes. Eight grasses and one legume have been compared for resistant qualities through two generations of the migratory grasshopper. Grasshoppers started on lettuce and transferred to the test plant during the fourth instar resulted in sexually mature individuals which laid eggs for the continuation of the study. The number of egg pods laid per female ranged from 6.3 to 12.7 for the parent generation and from 4.3 to 8.2 for the F_1 generation. The weight gain per male ranged from .012 to .072 g for the parent generation and from .002 to 0.68 g for the F_1 generation. The weight gain per female ranged from .030 to .115 g for the parent generation and from .015 to .125 g for the F_1 generation. It is concluded from these studies that the main mechanism of resistance is antibiosis. Two species of grass, blue bunch wheatgrass and green needlegrass, appear to be the most resistant to the feeding.

H. Insect Vectors of Diseases

1. Red Clover Virus. At University Park, Pa., red clover plots growing under cages which excluded insects showed much less clover virus and stands were more persistent than unprotected plots.

2. Sitona and Root Rot in Alfalfa. In cooperation with the Crops Research Division, a study was made at University Park, Pa., to determine the inter-relationship of the clover root curculio and Fusarium wilt of alfalfa. Yield reduction in the first cutting was caused only by Fusarium wilt. Reductions in yield in the second and third cuttings were due to both Fusarium and clover root curculio. The fourth cutting was reduced only by the clover root curculio. Fusarium wilt was the greatest cause of dead plants but the curculio contributed significantly to the number of dead plants.

3. Legume Viruses. Research was continued in 1965-66 in Poland under PL 480 project, E21-ENT-9, on insect vectors of virus diseases of forage legumes. Six different strains of pea aphid transmitted yellow bean mosaic virus but the rate of transmission varied between strains. Both the green peach aphid and the pea aphid were able to transmit simultaneously the yellow bean mosaic and red clover vein mosaic viruses from red clover to crimson clover.

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PRODUCTION, HARVESTING AND FARM STORAGE STRUCTURES,
EQUIPMENT AND METHODS
Agricultural Engineering Research Division, ARS

Problem. Better methods, techniques, equipment and structures for use on the farm for producing, harvesting, storing, and the initial preparation of grain, seed and forage crops for market are needed to increase efficiency in the use of labor and equipment, preserve quality and prevent spoilage and damage from mechanical handling and insects. Electromagnetic energy has many established farm uses but research indicates other highly useful potential capabilities in farm production, such as attracting and killing insects and improving the germination of seeds. There is considerable present need for precise seedbed requirements for various crops in different areas of the country. The exact best placement for starter fertilizer is also unknown for a number of crops under different climatic and soil conditions. Every method to control or eradicate plant diseases, weeds, and insects requires some type of equipment. Effectiveness of the equipment necessary may be essential to the success of the method which is attempted or recommended. The cost of harvesting and farm handling of forage, grain and seed crops is the major expense of production. The supply and adequacy of manpower for these operations are becoming progressively less satisfactory, thus requiring additional research in this area. While considerable information has already been obtained for the development of processes such as drying and separation, basic and more precise information must be developed for these and other processes in order to achieve further progress. New trends in storage structures also require additional research in design and use.

USDA AND COOPERATIVE PROGRAM

The Department has a continuing long-term program involving agricultural engineers, statisticians, physicists, and architects engaged in both basic and applied research in this area. Cooperation is maintained with experiment stations, the farm equipment industry and other scientific disciplines in the Department. The total effort involves 17.1 scientific man-years. Of this number, 1.0 are devoted to the use of electrical energy, 1.7 to equipment for establishment of forages, 1.7 to weed control in corn and soybeans, 2.7 to insect control in grain, 0.8 to forage harvesting equipment, 1.5 to forage seed harvesting equipment, 3.3 to grain drying, 0.8 to forage processing, 2.5 to seed cleaning and 1.1 to crop storage structures. Research is carried on at Beltsville, Md., Athens, Experiment and Tifton, Ga., Ames, Iowa, Lincoln, Nebr., Corvallis, Ore., Lafayette, Ind., Bushland, Tex., and Manhattan, Kans.

PROGRAM OF STATE EXPERIMENT STATIONS

Many of the State agricultural experiment stations are engaged in both fundamental and applied research dealing with the development of new principles and the application of currently available knowledge to the problems concerned in soil-machine relationships in order to increase efficiency in crop production. These studies are concerned in the broadest sense with the development of theories, special devices, and laboratory and field tests to determine ways in which tractive and transport equipment, tillage tools and systems for their use might be improved.

Problems concerned with planting of the many sizes and shapes of seed together with the introduction of fertilizers are under attack by many of the State agricultural experiment stations. These studies are concerned with the development of new principles that can be used to meter and place seed which could lead to planter improvement. Similar investigations are in progress to develop satisfactory metering and placement devices for application of liquid as well as solid fertilizers. In both instances the principal objective is to provide the best possible means of seed and fertilizer placement which will assure healthy plant emergence with vigorous growth to maturity.

Both basic and applied research investigations which have been designed to discover and develop methods, techniques, and equipment for control of the many pests that attack grain and forage crops are in progress at the several agricultural experiment stations.

Most of the State agricultural experiment stations are engaged in some aspect of basic or applied research which is concerned with improving machines and methods for efficient harvesting and farm handling of grain and forage crops. Detailed investigations are in progress to develop reliable mechanical harvesting and handling equipment as well as ways in which improvements might be made in crop production systems to increase yields, product quality and overall efficiency.

Many freshly harvested agricultural crops must be subjected to early treatment in order that they may retain as much as possible of their original qualities. The State agricultural experiment stations are involved in both basic and applied research studies which have as their broad objectives the development of improved methods, equipment and techniques for preparation and processing of farm crops to preserve quality and prevent spoiling while in storage.

The complicated problems associated with providing protection to the products of agricultural production as well as the machines, equipment and service facilities which are required for such production has necessitated

a continuing program of research at the State agricultural experiment stations. The current program is concerned with conditioning and storages for high moisture grains; structural characteristics, wall pressures, design and construction of silos; controlled atmosphere storages and construction methods; and designs and construction of plant growth chambers and plastic greenhouses.

Several of the States are engaged in programs of basic and applied research on the possible use of some of the various forms of electrical and physical energies as a means for improvement of the potential capabilities in farm production. Investigations in progress include the evaluation of the use of radiofrequency energy for treatment of grains to destroy insect infestation and treatment of seeds to improve their germination characteristics.

The agricultural experiment stations of many of the States have research under way whose major objectives involve the obtaining of information on the uses to be made of electrical energy to reduce labor, increase production and improve family living conditions. In the design of these studies provision has been made to develop and investigate new equipment and explore the possibilities for new uses for electricity on the farm. Many of the projects are concerned with the varied problems of chore labor mechanization and an expansion of the use of electricity for ventilating, heating, lighting and cooling under the various production enterprises of today's farming operations. Development and testing of prototype specialized equipment for product collection, processing, packaging, and transport, as well as crop storage, loading devices, are a part of the overall program of investigations.

The State agricultural experiment stations are engaged in extensive basic and applied research to extend the advantages of controlled environment to all phases of agriculture in order to obtain maximum economic growth, production, product preservation and product quality. Studies of the possibilities for use of solar energy as well as electric energy to achieve the broad scale objectives are a part of the total program. Among the several investigations involved in these programs are determination of the effects that soil, light and atmospheric conditions on plants; and temperature, humidity and gases on stored products. Special attention is being given to development of means for collection, storage and use of solar energy for crop conditioning.

Much of this research is cooperative with the Department.

PROGRESS -- USDA AND COOPERATIVE PROGRAM

A. Systems of Equipment for Conservation Farming

Studies were initiated in cooperation with the Iowa station to evaluate the use of herbicides and tillage practices for 30-inch-row corn production. Overall spray applications of atrazine, simazine, and 2,4-D were made in early spring on unplowed and spring-plowed cornstalk ground. Secondary tillage operations included disking, strip rotary till planting, and no tillage. Adequate rates of fertilizer were applied so that fertility level could be eliminated as a variable. The control or check treatment to which all others were compared was conventional moldboard plowing, disking, and spike tooth harrowing. Mechanical cultivations were performed as weed infestations demanded. Because of adverse weather conditions during the month of May, it was impossible to perform the secondary tillage operations or to plant until early June. This resulted in variable stands and low yields. Because of this, it was impossible to draw specific conclusions from this experiment. There were, however, some trends in the data that are of interest. Early applications of simazine and atrazine gave substantially better weed control with only one mechanical cultivation than the unsprayed plots or the plots that received 2,4-D as a spray. In all instances, the lowest stands, poorest weed control, and lowest yields occurred where no secondary tillage was performed. Previous work had indicated that adequate weed control was obtained without secondary tillage. The results this year indicate a need for secondary tillage not only from the standpoint of establishing adequate stand but also for obtaining better weed control. The trends indicate that corn can be produced in 30-inch rows with little or no seedbed preparation if adequate stands and weed control are obtained.

Studies were initiated in Iowa to compare ridge and conventional planting of corn and soybeans using 30-inch row spacings. Because of the narrower row spacing, the ridges were somewhat reduced in height and width. Faster emergence and equivalent yields were obtained when weeds were controlled chemically. Mechanical cultivations were somewhat more difficult for both ridge and flat planted corn and soybeans when the 30-inch row spacings were used. Chemical weed control appears to be more consistent with the ridge planting system.

Studies were initiated in Iowa to evaluate tillage systems under a high level of fertility and good weed control obtained through the use of preemergence applications of atrazine sprays in corn. The following systems were evaluated: (1) strip rotary tillage, (2) listing, (3) conventional disking and harrowing, (4) ridging, (5) Missouri till planter, (6) Dempster till planter, (7) dragging hoe, (8) disking, (9) strip tillage with a cultivator sweep, (10) no tillage. These systems were evaluated on fall plowed land. Adverse weather conditions made it necessary to delay the

secondary tillage operations until early June. This resulted in a low level of corn production and makes the results somewhat tentative. Trends in the data indicate that the omission of tillage resulted in fewer stalks per acre, a heavier weed infestation, and lower yields. Among the tillage systems, the yields, stands, and weed weight differences were quite small, indicating that all performed equally well. These results indicate that the tillage systems under test will probably be successful on 30-inch corn in the same manner that they were successful on 40-inch corn.

In cooperation with the Iowa station, the model tillage laboratory was used to study the influence of a simple oscillating tillage tool on the formation of soil clods. Clod size determinations were made before and after the tillage operations. Preliminary results indicate that the forces, velocities, and energy inputs along with the physical properties of soil can be correlated with the mean weight diameter of the resulting clods. It appears that the total energy inputs are somewhat reduced by oscillations.

B. Fertilizer Placement and Distribution Equipment

Eleven field experiments on the establishment of field crops and vegetables were put in cooperatively with various State and Federal research units. These experiments were put in at eight locations, which are within the bounds of six states (Florida, Georgia, Maryland, Nevada, Texas and Washington). Some typical results or trends shown of the field crops are: the yield of grain on winter oats at Watkinsville, Georgia, with 80 lbs. per acre of seed drilled in four inch rows with fertilizer placed one inch to the side and one inch below the seed row was increased about 30 percent over a similar seeding in eight inch rows. Another seeding drilled in four inch rows, but with the fertilizer placed in contact with the seed, produced grain yields about 19 percent greater than a similar seeding in eight inch rows. The recommended fertilizer used was 600 lbs. of 5-10-15 per acre.

C. Equipment for Establishment of Forages

1. Thirty-two field experiments on the establishment and production of pastures and haylands were put in or continued from the previous year in cooperation with various State and Federal research units. These experiments were put in at eleven locations, which are within the bounds of eight states (Georgia, Louisiana, Maryland, New Mexico, Oklahoma, Tennessee, Texas and Washington). Some typical results or trends shown of the forage studies are: interseeding of high quality grass into stands of low quality plants with minimum disturbance has been conducted at Pantex, Texas in 1963-65. The late May date of seeding appears to be the best date of seeding on the Texas High Plains. March and April plantings generally have fewer grass seedlings and more weeds than later plantings. This work will be summarized

for a publication in 1966. The influence of soil compaction on grass establishment has been studied at Big Spring, Texas, from 1963-65. The comparison of bulk density in 1965 with measurements made in 1963 indicate that the low soil densities are becoming more dense and the high densities becoming less dense. The field work on this experiment is scheduled to continue in 1966; however, since the Big Spring Field Station is scheduled to be discontinued in July 1966, the experiment probably will then be terminated. Plant materials, fertilizers, and cultural practices in relation to stabilization of sand dunes have been studied from 1963-65 near Guymon, Oklahoma. The most promising economical approach to stabilizing the dunes is to encourage growth of existing species through fertilization. Analysis and summary of this work are scheduled for 1966. From investigations cooperative with the Georgia station, it was found that the use of herbicides in the sprigging of Coastal bermudagrass is quite important -- is much more important when ample moisture is available the first few weeks of establishment than in seasons with little moisture (1964 experiment had no moisture the first six weeks -- 1965 experiment had good moisture up to first forage harvest in October). Water with sprigs as set (irrespective of herbicides) gave best initial stands, and survival stands (through first winter) of all methods when season moisture was poor. With ample moisture, establishment using water with sprigs was about equal to water applied as irrigation. The common method of applying fertilizer only after the sprigs show life gave poorest initial and survival stands during both seasons. With two more or less exploratory seasons completed, and trends showing considerable differences between good and poor moisture seasons, a new means will be used for the next season to get a series of tests under limited moisture conditions. An area 120 ft. x 24 ft. is being provided with removable covers, to be used for field plot experiments with controlled moisture. It is expected this new control will give results under adverse growing conditions much quicker and will provide more comparable data and, therefore, more reliable results.

In Western Washington phosphorus fertilizer broadcast on forage plots remained concentrated in the surface inch of soil, while potassium moved appreciably downward into the root zone. This localization of phosphorus has also been noted in other humid areas. Phosphorus must be applied near the seed or feeder roots for maximum efficiency.

2. Work has continued under a contract with the New Mexico station to develop machinery to seed rangeland. Most of the work was accomplished at the Jornada Experimental Range, New Mexico, on sites where creosotebush and tarbush are the major types of undesirable vegetation. The primary objective of the research is to modify existing equipment, or to design equipment that will accomplish the following four operations in one pass over the land: eradicate undesirable shrubs, provide a firm seedbed, place seed accurately and windrow uprooted brush over the planted strip.

Three basic units have been used thus far in the investigation to accomplish the above objectives: (a) a rootplow to eradicate brush, (b) a seeder to firm the seedbed behind the rootplow and accurately place grass seeds, and (c) windrowing equipment to place uprooted brush over the planted strip to protect emerging plants.

Two basic units of the equipment remain unchanged from the previous year: (a) The Fleco Rootplow for brush eradication, and (b) the "New Mexico" Nisbet Range Seeder for firming the seedbed and planting the seeds. A D-7 caterpillar tractor is the source of power for the operation. During the period of this report, two models of a side-delivery windrower have been constructed, and a baled hay loader has been modified to investigate the feasibility of carrying the uprooted brush over the top of the seeder and depositing the residue immediately behind.

The following problems were encountered with the first model of side delivery rake tested: the tines did not have sufficient strength and several were deformed or broken during field trials, and the rotational acceleration and deceleration of the carts as they passed around each of the four sprockets at the corners caused failure due to fatigue in the connectors between chain and carts, and in the bolts which attached the carts to the connectors. Changes considered necessary have been made in the first model of the side delivery rake, and a second model is now ready for testing.

A traction-powered, baled hay loader was modified, under the New Mexico contract, to pick up the uprooted brush and elevate it to a height sufficient to drop it behind the Nisbet seeder. The modification consisted of attaching two 7-inch x 3/8-inch round steel rods to each of the lugs on the two hook chains of the loading mechanism, and removing the narrow, vertical side boards along the elevator platform and replacing them with wide sheet metal "wings" at a 30° angle from the horizontal. Preliminary tests have indicated the feasibility of moving the brush with the baled hay loader. The 7-inch spikes on the chain appear to be of sufficient length to pick up the brush and carry it up the platform inclined at approximately 40° with the horizontal. Other tests will be made using steeper slopes until the maximum slope is determined at which the brush can be carried up the incline.

D. Insect Control in Grain

1. Twelve granular insecticides or insecticide combinations were applied, in research cooperative with the Ohio Center, to corn plots for control of Northern Corn Rootworm within two days after the corn was planted. Where rootworm population was low, there was little increase in yield. Root damage and lodging were slight. In one plot series where the average larvae

population was twelve per plant in untreated corn, five insecticides (N2790, carbophenothion, GC 4072, dieldrin, Parahep) reduced lodging and produced higher yields (increase 18-34 bushel per acre).

An outbreak of chinch bug on corn occurred in June, in Ohio. Granular phorate (Thimet), sprays of malathion, and dimethoate were applied for experimental control. Phorate produced excellent control and either of the other two materials could be used for emergency control but some reinfestation could be expected to occur.

Field plot screening tests were continued, in cooperation with the Iowa station, for first and second brood borer control using both the liquid and granular formulations of a number of experimental and commercially available compounds. Although the organo-phosphates are somewhat more dangerous to apply, borer control obtained with such compounds as Parathion, Diazinon, etc., was equivalent to that obtained with DDT. The plot application equipment developed for the corn borer work was able to handle the materials without any serious problems. Conventional sprayers and granular applicators were modified and mounted on high-clearance equipment for this work. It is apparent that additional safety measures and possibly equipment changes will be needed for handling the more toxic compounds.

Studies were continued on the use of systemic insecticides for controlling first-generation corn borer larvae in corn. Four compounds, American Cyanamid 47470, General Chemical 6506, Union Carbide 21149, and Thiochron were used in these studies. These compounds were applied in the granular form at planting time, with the fertilizer, with the seed, two inches to the side of the seed, and two inches to the side and two inches below the seed. Some of these compounds were also applied at the base of the plant during first cultivation. Of the compounds tested, only #47470 gave effective control. The differences in control among the various placements tried were not very large. However, there was some indication that the chemicals applied with the seed caused some phytotoxicity and that placement two inches to the side and below the seed gave slightly poorer control. The systemics applied at cultivation time were not quite as effective as those applied at planting time. Equipment for applying these compounds was designed, developed, constructed, and calibrated specifically for these studies.

A series of tests were conducted on the use of granular insecticides to control both the first-generation corn borer and the corn rootworm. A total of nine experiments were conducted in seven locations in the western part of Iowa. Parathion, diazinon, Thimet and DDT were the compounds used. All fields were treated twice. The first application was made when evidence of rootworm feeding was found. The second application was made one week later. In one experiment, four levels of soil coverage were tried. These levels were one-half inch, one and a half inches, two and one-half inches, and three and one-half inches. In two of the experiments no cultivation was compared with the cultivation that resulted in a two and one-half inch soil coverage. The results show excellent control of both the corn borers

and the rootworms when the organo-phosphates were used. DDT gave excellent control of corn borers but inadequate control of rootworms. Diazinon gave slightly better control than the other compounds. The differences in control among the different levels of soil coverage were not large enough for significance, indicating a lack of importance for specific amounts of soil coverage level. The comparisons of cultivating and not cultivating showed some improvement where the chemicals were cultivated in; however, this improvement did not occur when moderate to heavy amounts of rain fell shortly after application. The differences were quite large and in favor of incorporation under dry conditions. These results indicate that both rootworms and borers can be controlled with one chemical.

Low volume applications of malathion with aircraft using the mini-spin nozzle and conventional flat fan-type nozzles were studied for controlling first-generation borers. A series of applications were made at four day intervals at three different locations. In addition to borer counts to evaluate control, spray droplets were collected on glass plates to study the spray droplet distributions. Borer control, as indicated by borer and cavity counts, showed little or no control with any of the low volume applications of malathion. The mini-spin nozzle produced fewer large droplets than the conventional fan-type nozzle. However, there was excessive drift with both nozzle types. In one instance, with a crosswind of less than five miles an hour, the spray swath shifted in the direction of the wind more than 100 feet. These results indicate very little if any potential for low volume aircraft applications of compounds like malathion for controlling corn borers.

Granular, emulsifiable, and in-capsule formulations of bacillus thuringiensis were applied for first-generation borer control. Results showed excellent borer control. These materials were applied with the field plot equipment used for the insecticide work. There were some problems with the emulsifiable formulations staying in suspension and with the flowability of the granular formulations. It appeared that some minor changes in the formulation processes would eliminate most of these problems and that the conventional insecticide application equipment could be used to make these applications.

2. A study was made in Georgia to develop equipment for applying either technical or concentrated materials at extremely low volumes. A low volume sprayer was designed and built which used either a flat-fan type or a pneumatic nozzle. The flat-fan type nozzle applied one quart per acre and the pneumatic nozzle applied one pint per acre. Results from these tests shows a significant difference between treatments; however, neither treatment gave adequate insect control when the equipment was operated with one nozzle above the middle of two rows. The equipment was redesigned and the nozzle was placed equi-distance between the rows at ear height. These changes were made too late in the growing season to adequately evaluate the the equipment. This sprayer appears suitable for applying low volume insecticides for grain or other crops where low volume applications may be effective.

A test was made in Georgia to determine whether insecticide and application procedures generally recommended for corn earworm control were adequate on late planted sweet corn. Three plots were used in this series of tests. One plot was treated every day, another every second day, and another every third day. Data from these tests showed that plots treated every day, every second day, and every third day had 41.7, 21.6 and 0.03 percent undamaged ears, respectively. The more frequent treatment was helpful in preventing insect damage, but none of the treatments gave satisfactory insect protection on this corn.

3. Several new methods were tried in Georgia for repelling corn earworms without the use of insecticides. Methods tried included: the use of vapor type street lighting lamps, and the use of high frequency sound. However, no control was obtained by the methods tried.

Corn was planted in Georgia with the seeds oriented with respect to the direction of the row, to study the effect on leaf distribution and consequently the effectiveness of spraying. While there was some difference in the leaf distribution because of orientation of the seeds while planting, there was no significant difference in insect control or yield.

E. Weed Control in Corn

1. Field studies were continued, in cooperation with the Missouri station, to evaluate different methods and equipment for directed applications of dalapon (one and two pounds per acre) for weed control in corn. The directed applications were made so that various methods of corn leaf protection could be evaluated. This protection varied from no leaf protection to a maximum protection provided by tying the leaves to the corn stalk. A special shield leaf lifter, wire leaf lifter, and directed nozzles were used on the sprayer. Applications were made under weed-free conditions so corn damage from contact with the dalapon could be evaluated. No significant damage due to any of the treatments was found. Corn stand and yield were not reduced when dalapon was directed onto the lower three to four inches of the corn stalk. The corn was allowed to grow more prior to application than in previous years which could account for some of the resistance to dalapon injury. It was first thought that injury was found in previous years when rainfall occurred after application, but this year 1.12 inches of rainfall occurred during the week after application. It has been determined from the four years of study that the wire type leaf lifter is satisfactory for application of dalapon in corn.

Field studies were continued to determine the effectiveness of 8-, 10-, 12-, 14-, and 16 inch band applications of 2,4-D (one pound per acre), simazine (two pounds per acre), and atrazine (two pounds per acre) for weed control in corn. Herbicides applied in narrow bands result in a significant reduction in applied chemical. There was no significant reduction in corn yield due to decreasing the band width. No lateral movement of the herbicide was observed so weed control was limited to the applied area again

this year. Better overall weed control was obtained when the herbicides were applied in wider bands. This study will be continued another year.

Studies on weed control equipment and practices for narrow-row corn and soybean production, in cooperation with the Iowa station, indicate that the equipment and systems now used for conventional row spacing will give satisfactory weed control. These studies indicate a need for changing the cultivator sweep spacings for mechanical control and for changing the nozzle and granular spreading device spacings for chemical control. Earlier soil shading with the narrow rows resulted in over-all improved weed control at the end of the season.

Field scale studies were conducted in Iowa on soil incorporation with planter attachments using Ramrod, 2,4-D, Feniben, and amiben granules for weed control in corn. All compounds were applied in 14-inch bands at planting time with and without soil incorporation. Amiben granules resulted in some corn damage when incorporated; however, this damage did not have a serious effect on yields. Results indicate little or no advantage for soil incorporation of these compounds.

F. Forage Harvesting Equipment

1. Studies of the effect of methods of harvesting on processing energy requirements showed that field-wilting of Coastal bermudagrass greatly reduced the energy requirements for grinding and pelleting. The pellets produced in these studies were fed to steers and the results of feeding are not yet available. From the chemical analysis, however, it does not appear that the hay was appreciably reduced in quality by the field-wilting.

2. A new hay wafering process and the resulting product was evaluated. The wafers were formed by rolling the hay into cylindrical units. The experimental machine was small, lightweight, and simple in design. A basic problem existed in feeding the hay into the wafering device so that the latter worked properly. Unlike extrusion wafering, high moisture forage and forage consisting of grasses could be wafered by this rolling or wrapping process without the addition of moisture and with relatively low power consumption. The experimental wafers were less dense than most formed by extrusion, but equally durable. The wafered hay was not free-flowing. It could be satisfactorily dried though the wafers released moisture slowly. The rolled wafers were eaten satisfactorily by dairy cattle.

3. Significant field losses occurred from raking forage regardless of how damp the forage. The least field losses occurred by cutting and windrowing the forage in the same operation with a self-propelled windrower and handling the windrows with a windrow turner. Slightly more alfalfa was stored in a tower silo as haylage than as direct-cut silage. A slight increase in density occurred when dewatered forage was ensiled, however, the processing

and ensiling losses were as great as those from ensiling direct-cut silage. The most dense silage was only about half as dense as the maximum possible if the voids could be eliminated. Storing forage as haylage resulted in the least losses.

G. Forage Seed Harvesting Equipment

1. Development of a high-speed scalper for seed crops. Several kinds of rotating scalpings were designed, constructed, and tested in an effort to develop a machine to separate straw from threshed seed in a combine. Rotating fingers, squares, and other shapes were tested, but the separating efficiency was not comparable to the present method.

2. Development of components for cutting, picking up, threshing and cleaning field seed crops. Rotary cutter tests were continued in 1965 to develop a new method of picking up seed crops for an improved-type combine. A rotary cutter, with slight alteration, was again tested in several grass and legume crops. Total seed picked up was only 39.7 percent for windrowed red clover, 42.1 percent for standing red clover, 53.1 percent for windrowed creeping red fescue, and 60.5 percent for windrowed Newport bluegrass. These results are not adequate; therefore, the machine will be completely remodeled and tested again during the coming year.

A belt thresher is being developed that will provide a more efficient threshing unit for an improved-type grass and legume combine. The experimental machine, as constructed, employs the same threshing principle as the laboratory rubber-covered rub board. Therefore, it is expected to handle difficult-to-thresh and easily-damaged seeds without injury. Unthreshed seed fed between rough surfaced belts is rubbed and rolled by the belts traveling at different speeds until the seed is threshed. Preliminary tests indicate that with proper adjustment of feed rate, belt speed, and belt pressure, crimson clover can be satisfactorily threshed.

Time-of-harvest studies were continued on bluegrass in 1965, for a fourth year, to determine the optimum mowing time as indicated by pure-live-seed yield. Both Merion and Newport varieties were mowed and windrowed six times over a 23-day period starting at a seed moisture of 50 percent for Merion and 45 percent for Newport. Again, seed color, shatter, and other criteria were found to be unreliable indicators of optimum maturity and cutting time. The optimum time to harvest both varieties of bluegrass was found to be at the first mowing and when seed moisture was highest. Since the optimum time was earlier in 1965 than the time that the crop had been harvested in any previous year, the study will be continued in 1966 and the test will start early enough to define the highest point on the pure-live-seed-yield curve.

Time-of-harvest studies were started on fineleaf fescue in 1965 to determine the optimum mowing time as indicated by pure-live-seed yield. Both creeping red and Chewings varieties were mowed and windrowed six times over a 35-day period starting at a seed moisture of 59 percent for creeping red and 57 percent for Chewings. The optimum time to harvest both varieties of fescue was found to be when seed moisture was 23 percent. This harvest date was 7 days after farmers mowed in the same field, and resulted in a 15 percent increase in pure live seed. Since this is the first year's study for fine-leaf fescue, it will be continued at least one more year.

Seed moistures for all crops were again checked by both the exhaust oven moisture tester and the standard electric oven method. Readings with the exhaust meter were within ± 2 percent of the electric oven readings with a majority falling with ± 1 percent.

H. Seed Cleaning

1. Seed cleaning research applied to specific problem mixtures. Seed cleaning research was continued in the attempt to improve upon existing techniques for processing given seed mixtures--either by performing a more precise separation with conventional equipment or by developing and using new equipment to better exploit seed differences. The recurring problem of separating various contaminants from bentgrass was again considered. Dirt clods were removed effectively with a gravity table and a special indent cylinder. Mouse-ear and other short weeds also responded well to the special indent. A different contaminant--spike bentgrass--could not be removed satisfactorily even by extensive trials with the pneumatic, velvet roll, vibrator, and gravity (sandpaper deck) separators. Seed measuring and screening trials were carried out to determine the best screen and indent sizes for separation of certain seed mixtures. Separating results predicted from seed measurements were then correlated with actual screening tests. Compilations of seed measurement data were processed to obtain arithmetic means and standard deviations for length, width, and thickness dimensions. These values were established for Kentucky bluegrass, Colonial bentgrass, alfalfa, and cotton. In equipment development, the single-deck vibrator separator continued to demonstrate its versatility by removing various weed seeds from white clover, carrot, alfalfa, orchardgrass, and Rhodes grass. Brief trials with the resilience separator showed that quantity and purity of final fractions from a seed lot could be varied by moving the bounce plates horizontally or vertically. In separator testing, the magnetic process was investigated to learn how separation efficiency is influenced by amount of moistening liquid, types of metal power, and additives like soluble oil and wetting agents. These variables were evaluated in different test combinations by determining how effectively dodder and buckhorn plantain were removed from red clover.

In general, the amount of liquid was found important but liquid additives were relatively unimportant. About one-third pound of water per bushel of seed was optimum. Metal type also was important, with the finer metals doing the best job of removing weed seeds. In testing the color separator, various contaminants were removed from beans, rice, mustard, onion, safflower, barley, and soybeans. Other brief trials were conducted to learn how a given product responded to different arbitrary combinations of colored filters and background colors. In pneumatic separator tests, the selectivities of five machines (with various modifications) were compared by separating standard test lots at two air velocity ranges. By analyzing the relationship between material lifted and lifting-air velocities, numerical rankings were established for individual machines or modifications within batch or continuous flow groups. In addition, batch separators as a group were found more selective than continuous units. Round and square column shapes showed about equal selectivity in batch tests, but were far exceeded by a zigzag column. Given separators changed ranking spots when tested at both low and high velocity. Finally, the selectivity of a separator tended to decrease as terminal velocity of the product handled increased.

2. Development and testing of the centrifugal screen separator was continued during 1965. A number of mechanical changes were made that provided considerable improvement in the smoothness of operation and in mechanical dependability. Additional study was devoted to possible designs for a new model with mechanical changes which would enable the new machine to be used as a component of the separator section of a combine. This development could revolutionize the separation of granular material that would increase both machine capacity and efficiency, resulting in a higher quality product at a reduced cost.

I. Grain Drying

1. At Ames, Iowa, studies are underway for isolating the factors that enter into rational design of drying equipment and develop quantitative descriptions of their relation to economic design. Measurements of the rate of moisture loss from thin layers of kernels are made to determine the separate effects of air temperature, air humidity, air velocity, initial grain moisture, initial grain temperature, and other grain properties on moisture movement. In exposed kernel drying, about 50 grams of kernels are arranged in a single layer for air to pass through. The weight is observed periodically to observe the rate of loss of moisture. With soybeans, a consistent pattern is observed at all conditions of initial moisture, air velocity, air temperature and humidity, initial kernel moisture, and initial kernel temperature. The consistency is such that numerical coefficients can be assigned to describe the effect of each. The coefficient for velocity is poorly defined; partly because of the difficulty of controlling and

measuring velocity in the vicinity of the kernels. High velocity has the effect of speeding the drying during the early stages but has no observable effect on the drying rate later in the drying period. The effect of initial grain temperature was also poorly defined. If the grain is cold when the test is started, drying is slowed for the first few minutes. In some cases the moisture content may actually increase before any moisture is lost. The net effect of low initial temperature is to delay the drying so that it takes longer to reach any given level of moisture. In these tests the delay ranged from 2 to about 10 minutes. The numerical coefficients describing the effects of the other factors repeat themselves pretty well from test to test.

Earlier tests on counterflow drying of corn have been analyzed more completely to yield design data for practical counterflow drying equipment. These results show how the necessary drying time is related to depth of grain bed and how much increase in drying capacity can be accomplished by the sacrifice of given percentages of fuel use efficiency.

2. Studies of the drying zone in mechanical grain driers were continued through use of laboratory model drying bins. The drying zone refers to that part of the grain in a drying bin in which drying is actually taking place at any time. The zone moves through the grain but, in general at any time during drying, part of the grain is drying, part is already dry, and part is still at its initial moisture. The temperature and moisture gradients in the drying zone determine the efficiency and the required drying time in any drier. Accurate definition of the drying zone in terms of grain moisture, air temperature and humidity, rate of airflow, etc. would permit rational design of drying equipment. While much progress has been made in relating these factors to the drying zone, drier design still remains an art without the support of adequate design data.

Very good data were obtained from the measurement of temperature with thermocouples spaced through the column of grain in model bin driers. Contrary to common expectation, it was found that the profile of temperature in the drying zone did not reach a fixed pattern and then proceed unchanged as the drying zone moved on through the rest of the grain. No static or steady state condition of temperature change was detected. In tests in which the drying air conditions were maintained constant, calculations of the amount of water removed from the grain based on temperature and airflow measurements agreed within 1 to 5 percent with measurements of water removed based on changes in weight of the grain. From this it can be concluded that the apparatus and techniques have been perfected to allow the collection of reliable data. The problem remains to generalize the data with a mathematical model.

3. Effects of mechanical damage to field-shelled corn. The effects of several different damage treatments to corn kernels on the deterioration rate of 28 percent moisture corn were studied. The general observation would be that damage to the embryo of the kernel is more serious than

damage to the endosperm. The results are expressed with relation to the deterioration rate of hand-shelled corn with little or no damage. An arbitrary figure of 1.0 was chosen to represent the deterioration rate of undamaged hand-shelled corn. Hand-shelled kernels sliced with a razor blade on the endosperm had a relative deterioration rate of about 1.2. Kernels with the crown cut away had a relative deterioration rate of 1.9. In contrast, kernels which were treated with impacting the embryo without rupturing the skin had a deterioration rate of 2.0. Those treated with puncturing the skin over the embryo with a pin had a relative rate of 2.2. Kernels in which the skin over the embryo was sliced with a razor blade had a relative rate of 2.4 and kernels in which the tip was cut off had a relative deterioration rate of 2.85. It was observed that the relative deterioration rate of whole sound kernels selected from a sample of corn that had been shelled in the field had the same rate of deterioration (2.0) as hand-shelled corn in which the embryo had been bruised with an impact tool. One major equipment manufacturer has started an active program of checking mechanical damage with their harvesting machines.

4. The time limitation on storing grain under any given condition is dictacted by grain deterioration which is caused primarily by the growth of molds and bacteria. Of secondary importance may be the respiration or growth of the seed itself. The factors which influence the rate of growth of the microflora are grain moisture, temperature, and the amount of physical damage of the grain. It is the purpose of this study to evaluate the influence of these factors on the rate of growth of the microflora and subsequently the rate of deterioration.

Improved techniques and apparatus allowed the measurement of carbon dioxide production from shelled corn with as little as 13 percent moisture. Carbon dioxide production was measured from samples of hand-shelled corn, field-shelled corn, and hand-shelled corn that had been treated with different types of damage. Most of the studies were made with corn at about 28 to 30 percent moisture content. Some samples of hand-shelled and field-shelled corn were studied at several moistures between 28 and 13 percent. All studies were carried out at 65° F. The relative rate of carbon dioxide production between field-shelled corn and hand-shelled corn was about 3 to 1 for all moistures. The differences found in the rate of carbon dioxide production as correlated with type of damage to the kernel indicate that damage to the embryo results in faster deterioration than damage to the endosperm. Traces of aflatoxin were found in some samples of corn from the samples on which carbon dioxide production was measured, but so far there is no way to tell whether temperature or moisture levels influence the aflatoxin production.

J. Forage Processing

1. Artificial drying energy requirement studies showed that the drying efficiency of a forage dehydrator drops rapidly as the moisture content of the incoming material is reduced. Equations were derived to show both the

energy required to evaporate 1 pound of water from hay with various initial moisture contents and the energy required to produce 1 ton of dry matter from hay with initial moisture content ranging from 10 to 85 percent.

The effect of unit processes on the economically important constituents in Coastal bermudagrass and millet showed the following: As the age of Coastal bermudagrass increases from 3 to 5 weeks, the percent of protein and the carotene decrease. With this decrease is also a decrease in moisture content and an accompanying decrease in processing energy requirements. As a result of these factors, the processing energy cost per pound of protein and per million units of carotene is relatively constant. The maximum production per acre of these two constituents was obtained from cutting every 4 weeks. Millet contains considerably more protein and carotene than Coastal bermudagrass. It also has a much higher moisture content and higher processing energy requirement, causing the processing unit cost for these ingredients to be approximately 50 percent higher than for Coastal bermudagrass.

A study of the effect of the addition of fines to clean pellets showed that, regardless of the initial bulk density of either the pellets or the fines, the bulk density was increased by the addition of as much as 15 percent of fines by weight.

Processing treatments were applied to Coastal bermudagrass to determine their effect on digestibility. Digestibility was evaluated by the nylon bag in fistulated steer method. Dry matter disappearance, compared to the control, ranged from negative values for some treatments to a 13 percent increase in others. The greatest increase was obtained from the poorest quality forage.

K. Crop Storage Structures.

1. Silo design criteria. In studies at Beltsville to determine relative effectiveness of gastight tower and bunker silos, farm size silos were filled with similar orchard grass and each was managed by recommended methods. Stored silage densities were nearly equal; feedable silage recoveries were essentially equal, about 92%. No spoilage was recovered from the gastight tower, but the bunker had 3.6% spoilage mostly at the juncture of plastic wall lining and top cover. The bunker had high peak temperatures, but dissipated less total heat than the gastight tower. Preservations of individual nutrients was essentially equal in the 2 silos, implying that temperature variations below 36° C do not affect nutrient loss.

Determination of forage density in normal storage conditions, using a radioisotope, continued at Beltsville, Maryland. In a tower silo filled with wilted alfalfa, density measurement by radioisotope showed that silage directly under the drop point was of nearly uniform density up to within 10 feet of the final surface, while at the opposite side of the silo, density began to decrease within 20 feet of the surface. This shows a packing effect of the dropping forage during filling, and a possibility of increasing overall density. For absolute measurement of density of farm products the radioisotope method has not proved practical as a research tool. A large enough radioactive source requires excessive shielding; instruments are costly and unstable.

Availability of wilted orchardgrass at Beltsville, Maryland, enabled obtaining of a few additional bunker silo wall pressure readings. These will be combined with previously obtained readings for analysis.

2. Wilted grass silage storage. In studies at Beltsville, to determine the effect of chop length on wilted grass silage, 2 farm size concrete silos were filled with similar crop and procedure, except that one mass was chopped short and the other long. Increased silo capacity due to short chopping was less than 5%. Both silage masses gave feedable silage recovery greater than 90% in spite of the forage being very dry, and no definite effect of chop length was found. However, top spoilage was greater in the long chop. Temperatures indicated similar fermentation processes in the 2 masses, but the long chop dissipated more energy as heat. Sealing of silo doors with plastic sheet was adequate for both chop lengths. A top sealing procedure which extended the plastic cover below the silage surface at the wall was effective but wasteful of labor compared to the usual procedure of turning the cover up at the wall.

In another experiment, a silo filled with alternate layers of long-and short-chopped wilted alfalfa showed no evidence of an effect of chop length on preservation efficiency. However, as a whole, this mass showed a phenomenon not previously associated with silage; A high degree of gas convection. It was evidenced by a marked drying of the central part from movement of water vapor to the wall, where extreme putrefaction resulted; by a uniform oxidized condition of the central part, rather than a spoilage front around points of air entry; and by a separation into 2 distinct hot and cold temperature regions with the hot region extending during storage. This phenomenon may be the most critical factor in non-gastight storage of drier forages.

3. Hay wafer storage. In a resumption of previous studies at Beltsville, Maryland, cylindrical rolled alfalfa wafers were tested for ease of drying in a dual purpose (drying and storing) deep bin. Drying rate was limited by moisture migration within the wafers and was too slow to prevent moldy centers in the wafers for typical conditions in farm driers. This wafer form had slightly better mass breakup characteristics for unloading from a deep bin than did previously tested extruded wafers. Mass air flow rate was slightly greater than that in extruded wafers. An eight-foot depth of cylindrical wafers gave a maximum pressure on vertical walls of 25 lb/sq. ft.

L. Electric Traps for Grain Insects

Light traps are useful in studies of the European corn borer and in control work for determining the time of emergence, the dates of maximum flight activity, the comparative activity from year to year, and the timing of insecticide applications. Light traps have been operated near Ames, Iowa, since 1952 in cooperation with the USDA European Corn Borer Investigations Laboratory and the Iowa Agricultural and Home Economics Experiment Station. Trap collections of the European corn borer were much lower in 1965 than in 1964, indicating the general reduction in infestations as compared to the previous year. The traps will be operated in 1966 with the same cooperators.

M. Electric Traps for Turf Insects

Cooperative studies with Purdue University were conducted at Lafayette, Indiana, to investigate the use of electric light traps for controlling the larger sod webworm, Crambus trisectus (Walker), in lawns. Traps were used both to indicate the seasonal abundance and for control purposes with promising results. Trap records from the past two years indicate that the larger sod webworm comprises 68 percent of the webworm population in areas sampled at Lafayette, and that about 70 percent of the webworm moths captured were females, which still carried eggs. Very few larvae or pupae were found within 75 feet of single-lamp blacklight traps used in the experimental installations. These studies will be continued using traps installed in the same lawns which were used in 1965. Blacklight survey traps have been used extensively during the past several years by PPC in detection and quarantine work with the European chafer. Use of traps for European chafer surveys was expanded this year, and additional areas of infestation were discovered in northwest Pennsylvania and northeast Ohio. Preliminary tests of electric traps as a possible control measure for the European chafer were begun in New York this year in cooperation with ENT, ARS.

N. Radiofrequency Treatment of Grain and Forage Seed

Previous studies have shown that all developmental stages of all stored-grain insects studied can be killed by exposure for a few seconds to RF fields of sufficient intensity, which do not damage wheat for germination or milling and baking purposes. On a practical scale, the electrical treatment would be more expensive than chemical insecticide controls.

Efforts to improve the efficiency of the method by applying the energy in high-intensity pulses were successful for one species but did not increase mortality of stored-grain insects generally for energy input comparable to unmodulated operation. A new pulse modulator has been designed and partially constructed to provide shorter pulses which may be more effective in providing control of the insects infesting grain.

In physiological studies, RF treatment of larvae of the yellow mealworm, Tenebrio molitor (L.), resulted in increased weight loss and oxygen consumption in the treated insects as compared to the untreated. Increased protein synthesis and catabolism of amino acids were also noted, and these disturbances did not appear to be completely explained by heating effects alone.

The relative dielectric properties of grain and insects influence the amount of energy each absorbs from the RF electric field. Since the dielectric properties are frequency-dependent, knowledge of these properties over a wide frequency range would be helpful in determining optimum frequency ranges for insect control purposes. Development of a method for determining these properties in the 50- to 250-MHz range is nearly completed and will provide data to supplement that already available for insects and grain in the 1- to 50-MHz range.

Earlier experiments have shown that RF electrical treatments are effective in reducing the percentage of hard seed and correspondingly increasing normal germination in alfalfa, red clover, ladino clover, and to a lesser degree, in sweetclover. Quality of RF-treated seed samples continued to hold up as well as untreated seed after 5 years in uncontrolled storage. RF treatment for hard-seed reduction was studied on 27 different alfalfa seed lots from Utah, Nevada, Oregon, and Idaho, which were selected for high hard-seed content. On the average, hard-seed percentage was reduced from 36 percent in the untreated samples to 12 percent by the RF electrical treatment. Infrared treatments for hard-seed reduction were also obtained on all of these seed lots for comparison, and infrared treatments were equally effective in lowering hard-seed content. In additional studies with sweetclover seed, neither one nor two additional RF treatments produced further hard-seed reduction on samples which did not respond effectively to the initial RF treatment to lower hard-seed content.

Limited work with several varieties of seed corn showed that germination and early growth of some varieties were accelerated. Results were evaluated by daily measurements of radicle and plumule development.

At Pullman, Washington, sweetclover seed samples at 11 different moisture contents between 1.63 and 8.15 percent were irradiated in the glow discharge at 12 different treatment levels to further define relationships between seed moisture content and reduction of hard-seed percentage achieved by electrical treatment. Some reduction in hard-seed content was obtained at moisture levels of 5 to 6 percent. As the moisture content was reduced, a greater reduction of hard-seed content was achieved over a wider range of treatment levels. At moisture contents above 6 percent, any reduction in hard seed was accompanied by an equivalent increase in the number of dead seeds.

Experiments to determine the morphological effects of glow-discharge radiation on alfalfa seeds were conducted in cooperation with the Department of Agronomy, Washington State University. Histological methods were not successful because satisfactory sectioning of mature seeds was not practical. The use of the X-ray contrast method to follow the entry of radio-opaque dyes into the seed indicated that, in the majority of cases, the dye was absorbed through the hilum area of the seed.

Studies will be continued to evaluate effectiveness of RF energy for improving germination and emergence of different types of seeds.

0. Electric Equipment for Soil Warming for Plant Growth

Investigations concerning evaluations of electric systems for soil warming were continued in Indiana, Minnesota, and Maryland. The objective of the research was to determine the requirements for installation and management of electric soil warming systems which will maintain suitable turf conditions for activities in critical-use areas during cold weather. Turf heating involves adding heat to the rootzone of turfgrass plants to keep the soil from freezing, keep the turf greener, promote new rootgrowth and blade extension, and help melt snow.

Based on this research program, specifications have been prepared for installing electric heating cables in the turf of the new Civic Center Busch Memorial Stadium at St. Louis and in the Air Force Academy Stadium at Colorado Springs.

The research program showed that the particular design for any installation will depend on the extent of and use for each turf area, the climatic location, the availability of electric energy and the grass species used. Both polyvinyl-chloride insulated nylon-jacketed cables and mineral-insulated copper-clad cables performed satisfactorily. Dense turf with medium thatch was approximately equal to a plastic cover in reducing heat loss.

The capacity of heating cable output/unit area necessary was shown to depend on the weather conditions at a particular location and the condition desired for the turf area. At Lafayette, Indiana, power densities of about 10 watts per square foot sustained Kentucky bluegrass in an active, but not vigorous, growth state throughout the winter. A density of about 5 watts per square foot was suitable to sustain a green, thawed Kentucky bluegrass turf. No advantage in growth appeared to result from applying heat at low rates for long periods of time when compared with high rates for short periods of time. Results were directly related to total energy added. Less temperature variation occurred when the heat was applied while the air was coldest and the incoming solar radiation minimum. Ryegrass, overseeded December 1, 1964, was 3 inches high by December 30 on heated soil covered by plastic, while unheated soil also covered by plastic, repeatedly heaved and thawed, and seed did not germinate. Existing bluegrass plants were overstimulated when temperatures about 50° F. were maintained at 1-inch depth. These plants were injured by frost during sudden severe cold weather.

Experimentation to study the potential use of electric heating cables under ornamental shrubs was continued cooperatively with the Purdue Department of Horticulture. The objective is to eliminate winterkill due to desiccation and thereby extend the growing period for certain temperature-critical evergreens.

Preliminary studies indicated that the need of supplemental heat for turf was marginal in climates similar to the Beltsville area.

Preliminary results in Minnesota indicate that a conditioning period (gradual reduction in heat) is required prior to removing heat from turfed areas during the cold season. The most severe treatment was heating for 5 weeks after transplanting and abruptly discontinuing heat. Sod with no conditioning period exhibited the poorest color, knitting, root development, and rhizome activity of all treatments. Continuous heat resulted in the maintenance of green color and active root and rhizome development. These are preliminary results and the experiment will require continuing evaluation.

P. Grain Drying Equipment

Plastic collapsible solar air heaters for drying grain are under development and are being tested for the third year. A standard 4-mil clear polyethylene film has weathered well, but some physical breakdown has appeared at certain stress points. Use of a new sun-resistant polyethylene film is being investigated.

The present collapsible plastic solar air heaters provided supplemental heating of air during 1965 fall grain drying tests. Solar energy conversion efficiency was comparable with the previous 2 years' records. Sorghum grain was dried from 26 to 28 percent moisture content to 13 percent or less. Weather conditions were not favorable for natural air drying of sorghum grain at this high moisture content.

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- Hansen, R. C., and Stewart, R. E. 1965. Energy-Size Reduction Relations in Agricultural Grain Comminution. Transactions of ASAE, Vol. 8, No. 2.
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GRAIN AND FORAGE CROPS RESEARCH

of the

UNITED STATES DEPARTMENT OF AGRICULTURE
and related work of the
State Agricultural Experiment Stations

Section B

This progress report of USDA and cooperative research is primarily a tool for use of scientists and administrators in program coordination, development and evaluation; and for use of advisory committees in program review and development of recommendations for future research programs.

The summaries of progress on USDA and cooperative research include some tentative results that have not been tested sufficiently to justify general release. Such findings, when adequately confirmed will be released promptly through established channels. Because of this, the report is not intended for publication and should not be referred to in literature citations. Copies are distributed only to members of Department staff, advisory committee members and others having a special interest in the development of public agricultural research programs.

This report also includes a list of publications reporting results of USDA and cooperative research issued during the last year. Current agricultural research findings are also published in the monthly USDA publications, Agricultural Research, Agricultural Marketing, and The Farm Index.

UNITED STATES DEPARTMENT OF AGRICULTURE
Washington, D. C.
December 15, 1966

APR 13 1967
CURRENT SERIAL RECORDS



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II. NUTRITION, CONSUMER AND INDUSTRIAL USE RESEARCH

CORN UTILIZATION - FOOD

Northern Utilization Research and Development Division, ARS

Problem. Utilization of corn in products for human consumption is an outlet of great economic importance. In 1964, U. S. per capita consumption of meal, cereal food, sirup, sugar, and starch obtained from corn totaled 28.3 pounds. If corn used for production of alcoholic beverages is included, this figure would be about 25 percent greater. In processing corn for food and beverage uses, corn oil is obtained as a valuable byproduct. Since 1964, annual production of corn oil has exceeded 400 million pounds. Except for foots and refining losses, all of this oil is consumed in food products, principally margarine and salad and cooking oils.

The need for research is encountered primarily in dry milling of corn where the yield and quality of fractions are important both economically and in terms of consumer acceptance and nutritive value of final products. Improvements are needed in milling techniques, especially for old and artificially dried corn, if optimum results are to be obtained. More information is needed on the composition of corn and corn fractions in order to identify and minimize losses of nutritionally important components that may occur during processing. Such investigations should result in cheaper and more nutritious products and therefore contribute to increased utilization.

In addition, these studies provide a foundation for research on composition, processing, and utilization of new strains of corn now being developed that have significantly higher nutritional value than does ordinary corn. Success in this development could make corn the world's No. 1 food grain. U. S. corn production, which is now 4 billion bushels annually, could be readily expanded to neutralize the present 1-billion-bushel annual world shortage of cereal foods. Since this new corn differs physically from ordinary corn, much effort will be needed to develop suitable milling methods and to provide the knowledge necessary for development of suitable food products to meet dietary needs of world populations.

It has recently been discovered that certain oilseeds and cereal grains, including corn, are subject to infection by molds that can produce toxic products. To provide safe food products, as well as to minimize economic losses, research is needed on the detection of these toxins; on their quantitative analytical determination; and on development of processing techniques for their detoxification or removal from corn.

USDA AND COOPERATIVE PROGRAMS

The Department conducts a continuing, long-range program of research involving analytical, organic, and physical chemists, biochemists, microbiologists, systematic biologists, and chemical engineers engaged in basic, applied, and developmental studies pertinent to utilization of corn in food.

The Federal scientific effort for research on food utilization of corn totals 4.8 scientist man-years. Of this number, 1.2 are devoted to chemical composition and physical properties; 2.2 to microbiology and toxicology; and 1.4 to technology - process and product development.

Research at Peoria, Illinois, on chemical composition and physical properties (1.2 scientist man-years) is concerned with lipids and proteins of the corn kernel. During the reporting period, studies on nonprotein nitrogen compounds of corn were completed.

Research at Peoria, Illinois, on microbiology and toxicology (2.0 scientist man-years) is devoted to studies on the production of mycotoxins by Aspergillus flavus and other molds. The work also includes a survey of the incidence of aflatoxin in commercial samples of various grains. A research contract in effect with the Agricultural Experiment Station, South Dakota State University, Brookings, South Dakota, provides for a survey of various species of Aspergilli to find and identify those producing toxic metabolites. A portion of this effort (.2 scientist man-year) is allocated to research on food uses of corn.

Research conducted at Peoria, Illinois, on technology - process and product development (1.1 scientist man-years) comprises investigations on corn dry milling. A grant has been made to Pennsylvania State University, University Park, Pennsylvania, for basic studies on the mechanical and viscoelastic properties of shelled corn as related to the corn dry-milling process (.3 scientist man-year).

PROGRAM OF STATE EXPERIMENT STATIONS

A total of 6.0 scientist man-years is devoted to this area of research.

PROGRESS -- USDA AND COOPERATIVE PROGRAMS

A. Chemical Composition and Physical Properties

1. Corn proteins, lipids and nonprotein nitrogen components. Compositional studies on corn are relevant both to food and feed utilization. Results are reported under Area 3, subheading A.

B. Microbiology and Toxicology

1. Aflatoxin investigations. Studies on toxins produced by molds are relevant both to food and feed utilization of corn. Results are reported under Area 3, subheading B-2.

C. Technology - Process and Product Development

1. Corn dry milling. Studies on corn dry milling showed that stress-crack formation increased as the initial moisture content of the corn decreased.

Stress-crack formation also was influenced by the temperature of tempering and took place most rapidly in the 65-85° F. range. Pretempering increased the yield of -4+6 grits by about 50 percent for two lots of corn dried under severest "dryeration" conditions. Improved results were obtained when the cold-tempering procedure was modified by adding the temper water in five rather than two steps. Results did not, however, equal those of pretempering or tempering with a steam-water mixture. Water flotation was found to be a promising means for removal of germ and some grit with attached germ from corn degerminator product in the -3½+25 mesh range.

2. Mechanical and viscoelastic properties of corn kernels. Research in progress under the grant to Pennsylvania State University showed that swelling stresses occurring in the corn kernel during moisture absorption can be interpreted by treating the kernel as a cellulosic gel. A number of methods and techniques developed for other gel structures, such as wood, presumably should be applicable to corn.

PUBLICATIONS -- USDA AND COOPERATIVE PROGRAMS

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Chemical Composition and Physical Properties

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*Research supported by PL 480 funds.

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CORN UTILIZATION - FEED
Northern Utilization Research and Development Division, ARS

Problem. Over 90 percent of the U. S. annual production of corn is used as animal feed. Corn is fed to animals in various forms including ear corn, shelled corn, cracked or ground corn and, in certain mixed feeds, corn gluten and other milling fractions. Because of the extremely large volume of this outlet, even small improvements in quality or processing efficiency are economically important to the feed industry and to the farmer.

The components responsible for certain nutritional effects attributed to corn, such as growth stimulation and improved feed utilization efficiency, have not been satisfactorily identified, nor are processing steps available that take these components into account. More information is needed generally on the nutritionally important components of corn and on the changes that occur in them during processing. Besides their activity as Vitamin A precursors, carotenes contribute desirable color to milk and the body fat of cattle. Xanthophyll pigments similarly impart yellow color to egg yolks and to the skin of broilers and fryers. Adequate information on these pigments and on their fate during processing is also needed to insure maintenance and improvement of quality.

Compositional and related processing research is required on several new strains of corn that are expected to become commercially important. These strains include (1) those expected to lead to corn capable of providing a balanced source of amino acids, and (2) those that contain increased amounts of xanthophyll and other carotenoid pigments and therefore would improve the competitive position of U. S. corn in world markets.

Another important direction for research is the fermentative conversion of corn grain and corn sugar to nutritionally important feed additives. The value of corn-based media for production of vitamins, β -carotene, and antibiotics is well known. However, possibilities for additional important developments are virtually unlimited and should be investigated on a continuing basis. Corn steep liquor is frequently used as a supplement in fermentation media and may also be added to feedstuffs. More information is needed on corn steep liquor to identify the factors responsible for its stimulating effects on growth.

It has recently been discovered that certain oilseeds and cereal grains, including corn, are subject to infection by molds that can produce toxic products. To provide safe feed products, as well as to minimize economic losses, research is needed on the detection of these toxins; on their quantitative analytical determination; and on development of processing techniques for their detoxification or removal from corn.

USDA AND COOPERATIVE PROGRAMS

The Department conducts a continuing, long-range program of research involving analytical, organic, and physical chemists, biochemists, microbiologists, systematic biologists, and chemical engineers engaged in basic, applied, and developmental studies pertinent to utilization of corn in feed.

The Federal scientific effort for research on utilization of corn in feeds totals 4.4 scientist man-years, of which 1.1 are devoted to chemical composition and physical properties and 3.3 to microbiology and toxicology.

Research at Peoria, Illinois, on chemical composition and physical properties (1.1 scientist man-years) involves study of lipids and proteins of corn kernels. During the year, research on corn carotenoid pigments and on nonprotein nitrogen compounds was completed.

Research at Peoria, Illinois, on microbiology and toxicology (2.0 scientist man-years) is concerned with studies on the production of mycotoxins by Aspergillus flavus and related molds. Research contracts (1.3 scientist man-years*) are in effect with A. D. Little, Inc., Cambridge, Massachusetts, for studies on stabilization of fermentative β -carotene; with Consolidated Laboratories, Inc., Chicago Heights, Illinois, for research on the use of antimetabolites to facilitate selection of higher yielding strains of microorganisms producing β -carotene; and with the Agricultural Experiment Station, South Dakota State University, Brookings, South Dakota, for survey of the genus Aspergillus to find and identify species producing toxic metabolites.

The Department also sponsors research in this area conducted under grants of PL 480 funds. Research on microbiology and toxicology involves a grant to the Agricultural University, Poznan, Poland, for studies to increase the yield of β -carotene produced by fermentation of cereal grains. Effort on this project is prorated among corn, wheat, and sorghum. During the reporting period, research was completed at the "Giuliana Ronzoni" Scientific Institute of Chemistry and Biochemistry, Milan, Italy, on production of Vitamin B₁₃ and at the National Institute for Agronomic Research, Paris, France, on mutation of yeasts for improved feeds.

PROGRAM OF STATE EXPERIMENT STATIONS

A total of 4.0 scientist man-years is devoted to this area of research.

PROGRESS -- USDA AND COOPERATIVE PROGRAMS

A. Chemical Composition and Physical Properties

1. Nonprotein nitrogen components. In addition to previously reported nucleotides, the following compounds from the whole mature kernel and endosperm were characterized: nicotinamide adenine dinucleotide, thiamine

*Work covers more than one commodity; only effort allocated to corn is included in total.

and uridine monophosphates, and guanine diphosphate. Two nucleotides containing thymine were found exclusively in the endosperm. Yields of nucleotides (weight basis) declined during maturation of corn. Larger contents of adenine and uridine nucleotides were found in immature corn, whereas cytosine and thymine nucleotides appeared only in the later stages of development. These studies, now completed, have been replaced by an investigation of the distribution, composition, structure, and properties of proteins from corn, including different genetic strains.

2. Carotenoid pigments of corn. Carotenoid pigments were determined in 575 corn samples furnished by Crops Research, but xanthophyll levels did not exceed the high value of 77 p.p.m. given in last year's report. These results confirm that breeding can raise the xanthophyll content of corn above the value of 60 p.p.m., a level at which 60 percent corn in poultry feed will provide a sufficient source of pigment for eggs. This research has been terminated.

3. Lipids in hybrid corn kernels. Preliminary studies have confirmed a satisfactory method for extracting "free" lipids from ground yellow corn and corn fractions. A thin-layer chromatographic technique appears to give adequate separation of the lipids into five classes: (1) unknown, probably includes phospholipids; (2) mono- and diglycerides or sterols; (3) free fatty acids; (4) triglycerides; and (5) hydrocarbons.

B. Microbiology and Toxicology

1. Microbial carotenoids. Studies on factors influencing biogenesis of xanthophylls in Chlorella variegatus provided evidence that these pigments are an integral part of the electron transport system. The concentration per cell cannot, therefore, be significantly changed. This research has been terminated.

Contract research by the A. D. Little Company showed that good stability could be achieved by extrusion of crude fermentation β -carotene with carriers such as gum arabic, carboxymethyl cellulose, and mixtures of dextrose with gelatin or starch. Reported stabilities included values as low as 77 percent after 5 months and as high as 100 percent after 7 months. At Consolidated Laboratories, Inc., a number of mutant strains of the β -carotene organism were isolated, but none gave higher yields than strains already available.

2. Aflatoxin investigations. In engineering studies all commitments for quantities of aflatoxin, either as whole metabolite or as a dry crude extract, have been met. The process used for production gave yields of aflatoxin B₁ exceeding 1 g./kg. of solid rice substrate. An extraction procedure was devised that gave a dry powder containing 70 percent of aflatoxin B₁. Products have been provided to the Western Division, to the Food and Drug Administration, and to the National Animal Disease Laboratory.

Strains of Aspergillus flavus were found that produce mainly aflatoxin G₁ on wheat and soybeans as substrates (ratio B₁:G₁ = 1:2). Aflatoxin was produced by three A. flavus strains when grown on oat hulls or groats. Production of aflatoxins at 11° C. by A. flavus was demonstrated.

Degradation of aflatoxin by 11 Pseudomonas spp. was found to be nonspecific. However, both growing and resting cells of Flavobacterium aurantiacum removed aflatoxin irreversibly from several test systems. Milk, corn oil, peanut butter, peanuts, and corn, artificially contaminated with aflatoxin for test purposes, were detoxified by addition of F. aurantiacum cells. Tests with ducklings showed that removal of aflatoxin by viable bacterial cells was complete and no new toxic products were formed. Contaminated soybeans were partially detoxified. Autoclaved cells and cell walls took up aflatoxin, but it could be eluted by repeated washing.

Forage crops (alfalfa, timothy, mixed red clover, sweet clover, and oat straw) were investigated as substrates for aflatoxin production. Toxic levels were produced on timothy, sweet clover, and oat straw by two strains of A. flavus. Aflatoxins B₁ and G₁ were found in all samples of spores of A. flavus NRRL 2999. Aflatoxin was found to reduce early growth of seeds (11 plant species) germinated in its presence.

Aflatoxin analysis was greatly expedited by development of an automated procedure for reading thin-layer chromatographic plates.

In the survey of commercial grains for incidence of aflatoxin, 1,054 samples of corn had been assayed as of June 1, 1966, with positive results (19 p.p.b. or less) for 34 samples. Of these, 24 were in Sample Grade, 5 in Grade 5, 2 in Grade 4, 1 in Grade 3, and 2 in Grade 2. For soybeans, 6 of 711 samples assayed gave positive results; 4 were in Sample Grade and 2 in Grade 4. One sample of soybeans gave an unequivocal positive result by chemical analysis. The other five contained small amounts (3-6 p.p.b.) of fluorescent factors that interfered with analysis. A combined extract from these five samples was toxic in biological tests with ducklings. Survey of corn and soybean samples is continuing.

Examination of wheat and sorghum samples has been completed. Of 534 samples of wheat assayed, 2 gave positive results; both were in Sample Grade. For sorghum, 6 of 533 samples assayed gave positive results; 4 were in Sample Grade and 2 in Grade 4.

About one-third of 175 samples of oats contained fluorescing factors that interfered with chemical analysis. Biological tests indicated absence of toxic material. An improved procedure which eliminated interfering substances has been devised and will be used in a new survey of oat samples.

Although conclusions should not be drawn until the entire survey has been completed, results so far are reassuring in that almost all of the positive responses were observed in the lowest grades of grain and no extremely high levels of aflatoxin were encountered.

Aflatoxin research has been augmented by a research contract with the University of South Dakota for survey of toxin formation by species of the genus Aspergillus.

3. Vitamin B₁₃. At the "Giuliana Ronzoni" Scientific Institute of Chemistry and Biochemistry, Milan, Italy, research has been completed under a PL 480 grant for studies on Vitamin B₁₃. Results showed that this growth factor is not a single compound as some believed. Instead, it is composed of a multiplicity of carboxylic acids, phenols, ketones, and cyclic peptides. Concentration of the growth activity is in the peptidic fraction. The individual cyclopeptides were separated, but their structures remain unknown.

4. Improved feeds by mutation of yeasts. Owing to resignation of key personnel, the National Institute of Agronomic Research, Paris, France, terminated prior to completion its research under a PL 480 grant for investigations of mutant yeasts capable of producing high yields of sulfur-containing amino acids. About 1,800 strains of yeasts will eventually be sent to the Northern Division.

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CORN UTILIZATION - INDUSTRIAL PRODUCTS
Northern Utilization Research and Development Division, ARS

Problem. Almost 2.5 billion pounds of cereal starches and flours are used annually in the U. S. for industrial purposes. Corn is the source of most of these products. Industrial outlets for starches and flours are, however, constantly threatened by synthetic products derived from non-agricultural sources. Maintenance of the present and future competitive position of corn starch and flour in industrial markets requires a continuing program of basic and applied research.

The most promising outlets for new and improved industrial products derived from corn include the paper industry, industrial chemicals, adhesives, protective coatings, plastics, elastomers, and thickening agents. The greatest opportunity exists in the manufacture of paper and paperboard products, the U. S. production of which is 43 million tons per year and growing at the rate of 3.3 percent per year as compared to GNP growth of 3.4 percent and population growth rate of 1.7 percent per year. Use of starches in paper doubled between 1950 and 1963 from 552 million pounds to 1.1 billion pounds. This growth rate is 5.6 percent per year. The average amount used per ton of paper product has increased from 19 to 29 pounds. This favorable picture stems from research conducted in the past. To maintain or, more desirably, to increase the utilization of corn starch and flour in competition with synthetics, new concepts must be evolved that relate chemical modification of starch with specific properties imparted to paper products. Technology must be developed to establish optimum procedures for industrial use of promising products currently under study such as starch xanthates, starch graft copolymers, cyanoethylated and sulfated starches, and the new starch from high-amylose corn.

At 5 to 6 cents per pound, corn starch is an attractive raw material for the manufacture of products that can find outlets in the multibillion-pound annual market for industrial chemicals. When fermentative, rather than conventional chemical, conversion is applicable, even cheaper sources of starch such as flour and ground whole grain can be used as the raw material. Over a billion pounds of corn sugar is used annually in the manufacture of such well-known industrial chemicals as sorbitol, mannitol, citric and gluconic acids, and methyl glucoside. Promising leads requiring research to ensure successful future developments include nitrogen, sulfur, and unsaturated derivatives of starch, vinyl glucosides, industrial enzymes, and enzymatic starch conversion products such as oligosaccharides, polyols, and glucosides.

Adhesives represent a field long dominated by starch, which accounts for nearly half of the annual 2-billion-pound market for these products. However, competition by synthetic resins is especially vigorous and effective because of the specialized properties required to achieve increased

production of products like corrugated container board on automatic machinery. The overall growth rate of adhesive consumption is almost 7 percent per year. Since starch is usually cheaper than synthetic adhesive resins, prospects are good for meeting the competition through research designed to improve viscosity properties, bond strength, tack, and drying time of starch-derived adhesives.

The remaining outlets--coatings, plastics, elastomers, and thickeners--represent a multibillion-pound market in which starch-derived products having suitable properties should find ready acceptance. Microbial polysaccharides, starch graft copolymers, and urethane foams from starch-derived polyols typify the products that result from research on corn starch and flour.

Research oriented specifically toward particular industrial applications of final products must be founded on a vigorous and wide-ranging program of basic and exploratory investigations. Such studies lead to the discovery of new concepts, principles, and reactions that are the source of new processes and products for future development.

USDA AND COOPERATIVE PROGRAMS

The Department conducts a continuing, long-range program of research involving analytical, organic, and physical chemists, biochemists, microbiologists, systematic biologists, and chemical engineers engaged in basic, applied, and developmental studies on the composition of corn, on characterization and properties of the components, and on their chemical and microbiological conversion to useful industrial products.

The Federal scientific effort for research on industrial utilization of corn totals 73.0 scientist man-years. Of this number, 12.2 are devoted to chemical composition, physical properties and structure; 25.2 to chemical and physical investigations to improve products; 21.4 to microbiology and fermentation; and 14.2 to technology - process and product development.

Research at Peoria, Illinois, on chemical composition, physical properties and structure (9.9 scientist man-years) involves study of starch, amylose, amylopectin, proteins, and lipids of corn. Much of the work is related to problems pertinent to high-amylose corn. A research contract at Arizona State University, Tempe, Arizona, for basic research on reactions of "V" amylose has been completed. Grants (2.3 scientist man-years) have been made to the Nebraska Agricultural Experiment Station, University of Nebraska, Lincoln, Nebraska, for basic studies on variations in starch granules of genetically different corn samples; to Princeton University, Princeton, New Jersey, for basic research on the relationship of viscoelastic properties of amylose film to structure and function of plasticizers;

to Iowa State University, Ames, Iowa, for basic research* on heat, mass, and momentum transport of cereal starches and flours; to Purdue Research Foundation, Lafayette, Indiana, for research* on the effects of disulfide bond cleavage on the structure of corn and wheat endosperm proteins; and to Arizona State University, Tempe, Arizona, for basic investigations of the helical structure of amylose.

Research at Peoria, Illinois, on chemical and physical investigations to improve products (19.1 scientist man-years) is directed to wide-ranging study of the chemical reactions of starch with the objective of discovering new chemical products and processes having potential for industrial use. During the year, one phase of this work involving study of possible means for preparing amino acid derivatives of starch was completed and replaced by research on synthesis of halogen derivatives of starch. Research contracts (2.6 scientist man-years*) are in effect with the Arizona Agricultural Experiment Station, University of Arizona, Tucson, Arizona, for basic studies on the reaction of acetylene with methyl glucoside; with The Johns Hopkins University, Baltimore, Maryland, for basic research on the reactions of starch in fluid dynamic media; to the University of Pittsburgh, Pittsburgh, Pennsylvania, for studies on dielectric activation of starch; with Southern Illinois University, Carbondale, Illinois, for investigations on synthesis of maltooligosaccharides; with Stanford Research Institute, Menlo Park, California, for research on graft copolymers of cereal starches with vinyl-type monomers; and with the Institute of Paper Chemistry, Appleton, Wisconsin, for investigation of physical chemical factors affecting retention and effectiveness of starch xanthates and xanthides in paper. Contract research was completed by Ohio State University, Columbus, Ohio, on synthesis of amino derivatives of starch and by the University of Arizona Agricultural Experiment Station, Tucson, Arizona, on the reaction of starch with mercaptans. Grants (3.5 scientist man-years*) have been made to Ohio State University Research Foundation, Columbus, Ohio, for basic research on the reaction of vinyl ethers with carbohydrates; to Ohio State University, Columbus, Ohio, for basic investigations of unsaturated and sulfur-containing carbohydrates and of the amination of starch; to Purdue Research Foundation, Lafayette, Indiana, for studies on sugars containing carbon-bound nitrogen, phosphorus and sulfur; and to the University of Arizona, Tucson, Arizona, for basic research on the reaction of starch with diepoxides.

Research on microbiology and fermentation conducted at Peoria, Illinois, (21.4 scientist man-years) includes studies on the use of microorganisms to convert cereal-based media to industrially useful products such as chemicals, enzymes, polymers, and biological insecticides. A large collection of pure cultures of industrially and agriculturally important microorganisms is maintained. The Pioneering Laboratory for Microbiological

*Work covers more than one commodity; only effort allocated to corn is included in total.

Chemistry conducts research on microbiological reactions and products. Investigations on biological insecticides for Japanese beetle and on other insect control agents is cooperative with Entomology Research Division and Plant Pest Control Division. Research on plant antibiotics involves cooperation with Crops Research Division. Research contracts (1.0 scientist man-year*) are in effect at Michigan State University, East Lansing, Michigan, for basic research on enzyme activity in sporulation; at Kansas State University, Manhattan, Kansas, for investigation of stabilization of vegetative cells of the pathogenic organisms; at the University of Minnesota, St. Paul, Minnesota, for fundamental studies on the transfer of genetic determinants of sporulation from one microorganism to another; at Baylor University, Houston, Texas, for investigation* of morphological changes involved in sporulation; and at the American Type Culture Collection, Rockville, Maryland, for studies on preservation of certain microorganisms for which lyophilization is ineffective. Contract research at the University of Illinois, Urbana, Illinois, on the applicability of a sporulation factor produced by bacteria to Japanese beetle pathogens has been completed. Grants (2.2 scientist man-years*) have been made to Cornell University, Ithaca, New York, for fundamental studies on biphasic fermentation; to the Nebraska Agricultural Experiment Station, University of Nebraska, Lincoln, Nebraska, for investigations on the nature of amylase enzymes; to Kansas State University, Manhattan, Kansas, for investigations on separation of enzymes and proteins by disc electrophoresis; to Iowa State University, Ames, Iowa, for investigation* of bacterial amylases and their action patterns; to the University of Wisconsin, Madison, Wisconsin, for studies on the fine structure of polysaccharide B-1973; and to the University of Arkansas, Fayetteville, Arkansas, for investigation of the mechanism of enzymatic hydrolysis of starch.

Research conducted at Peoria, Illinois, on technology - process and product development (9.2 scientist man-years) is concerned with detailed study and evaluation of starch derivatives having definite potential for industrial utilization and of processes for making them. Research contracts (5.0 scientist man-years*) are in effect with Stanford Research Institute, Menlo Park, California, for process development of selected starch graft copolymers; with Battelle Memorial Institute, Columbus, Ohio, for developmental research on starch and other cereal grain xanthides and for studies on starch derivatives for use as colloids in water-emulsion paints; with Western Michigan University, Kalamazoo, Michigan, for evaluation of modified cyanoethylated starches for applications in paper; with Archer Daniels Midland Company, Minneapolis, Minnesota, for investigations on the use of starch glycosides in coatings and plastics; and with University of Akron, Akron, Ohio, for evaluation of starch and starch derivatives as reinforcing agents for natural and synthetic rubber. During the year, contract research on evaluation of starch polyol urethane foams was completed by Archer Daniels Midland Company, Minneapolis, Minnesota.

*Work covers more than one commodity; only effort allocated to corn is included in total.

The Department also sponsors research on cereal starches, conducted by foreign institutions under grants of PL 480 funds.* Research on chemical composition, physical properties and structure involves grants to the University of London, London, England, for research on debranching enzymes and their use in studying the fine structure of starch components (5 years, 1963-1968); to the University of Osaka Prefecture, Sakai, Japan, for development of an analytical method for carbonyl groups in carbohydrates (4 years, 1964-1968); and to "Giuliana Ronzoni" Scientific Institute for Chemistry and Biochemistry, Milan, Italy, for research on glucopyranose rings in starches and dextrans (5 years, 1962-1967). During the year, research on corn zein was completed at the National Institute of Agronomic Research, Paris, France.

Research on chemical and physical investigations to improve products involves grants to Hebrew University, Jerusalem, Israel, for studies on starch vinyl and epoxide graft copolymers (4 years, 1963-1967); National Institute of Technology, Rio de Janeiro, Brazil, for research on phosphorus- and sulfur-containing cationic starches (5 years, 1962-1967); Ahmedabad Textile Industry's Research Association, Ahmedabad, India, for research on starch-gum copolymers prepared by codextrinization (5 years, 1963-1968), and for studies on preparation and characterization of hydroxyethyl ethers of cereal starches (5 years, 1965-1970); Academy of Sciences and Chemical Institute "Boris Kidric," Ljubljana, Yugoslavia, for studies on modification of starch by moisture and temperature treatments (5 years, 1964-1969); and Plastics Research Institute TNO, Delft, The Netherlands, for research on preparation of metal alkoxides of starch for use as intermediates in synthesis (5 years, 1964-1969); University of Edinburgh, Edinburgh, Scotland, for studies on the mechanism and structural changes involved in thermal, acid, and alkaline degradation of starches (5 years, 1964-1969); and to the Institute for Fibres and Forest Products Research, Jerusalem, Israel, for studies on the mechanism and products of mild oxidation of starch (5 years, 1963-1968). During the year, research was completed on fatty chemical derivatives of starch dextrans at the Institute of Industrial Chemistry, Bologna, Italy, and on changes induced in starch by gamma-irradiation at the National Institute of Agronomic Research, Paris, France.

Research on microbiology and fermentation involves grants to the University of Milan, Milan, Italy, for basic studies on the metabolic pathway to 5-ketogluconic acid in Acetobacter species (5 years, 1961-1966); University of Allahabad, Allahabad, India, for collection of new Mucorales species (5 years, 1961-1966), and studies on survival of lyophilized microorganisms (5 years, 1962-1967); University of Newcastle upon Tyne (formerly University of Durham), Newcastle upon Tyne, England, for investigations of sugar phosphate derivatives in molds (5 years, 1962-1967); Central Drug Research Institute, Lucknow, India, for studies on aerobic actinomycetes in India

*Effort prorated among corn, wheat, and grain sorghum.

to find new accessions for the ARS Culture Collection (5 years, 1965-1970); to the University of Liege, Liege, Belgium, for research to find lytic enzymes of microbial origin (5 years, 1964-1969); to the University of Lodz, Lodz, Poland, for research on the fermentative production of itatartaric acid (5 years, 1963-1968); University of Tokyo, Tokyo, Japan, for research on the fermentative production of D-tartaric acid (5 years, 1964-1969), and of mevalonic acid (3 years, 1965-1968); to the Institute of Biological Chemistry, University of Rome, Rome, Italy, for studies on the preparation and characterization of dextran derivatives (5 years, 1961-1966); to the National Sugar Institute, Kanpur, India, for research on isolation of natural polysaccharide gums (3 years, 1965-1968); and to the National Institute of Agronomic Investigations, Madrid, Spain, for study and collection of aerobic species of actinomycetes (4 years, 1965-1969).

PROGRAM OF STATE EXPERIMENT STATIONS

A total of 4.0 scientist man-years is devoted to this area of research.

PROGRESS -- USDA AND COOPERATIVE PROGRAMS

A. Chemical Composition, Physical Properties and Structure

1. High-amylose (HA) corn development. During the reporting year 12,517 amylose analyses were reported to cooperating corn breeders. To date, a total of 17 samples containing at least 85 percent apparent amylose have been found. Techniques for single-kernel analysis based on excision of only a small portion of the kernel were improved, and about 300 individual kernels were successfully analyzed by this method. Despite the damage caused by sampling, these kernels showed a high percentage of germination. Reports from breeders and millers indicate a sharply increased industrial demand for HA starch, presumably for uses other than coating glass fibers. It is thought that much of the increase is in response to demand from the paper industry. Another interesting report is that demand for Class 5 starch (50-60 percent amylose) continues despite availability of HA corn having higher amylose content. Apparently there is a real possibility for marketing a series of HA starches varying in amylose content to fill specific needs of industry.

2. Genetic control of starch properties. Research under the grant to the University of Nebraska showed clearly that specific recessive genes control properties of starch such as water absorption, viscosity, gelatinization temperature, and enzyme susceptibility. Starch associated with wx gene shows the highest water absorption, that with ae gene the lowest. The su2 gene control contributes low viscosity and a low temperature of initial starch gelatinization. The ae starches showed the lowest enzyme digestibility; digestibility was reduced by ae gene even when occurring in combination with other recessive genes which normally result in starch of high digestibility. A combination of su2 and wx resulted in a starch more

rapidly digested than starch from su₂ or wx acting singly. This study could provide the basis for breeding varieties of corn containing starch with solution properties meeting specific requirements.

3. Chemical studies on corn components. Basic investigations indicated that lithium salts, urea, and guanidinium salts disperse starch granules by production of a surrounding aqueous medium having a dielectric constant greater than that of water. "R-enzyme" as obtained from potato tubers was separated into two components. Approximate molecular weights of the components were 20,000 and 1,500. The high-molecular-weight component shows α -amylase activity, i.e., it hydrolyzes both amylose and amylopectin. The small molecule increases iodine affinity of the substrate apparently by increasing turbidity of the solution. Molybdate ion prevented precipitation of this small molecule and therefore repressed this reaction.

In the contract research at Arizona State University, now completed, more precise study of the reversible dehydration of V-amylose hydrate gave a value for ΔH of 10.4 kcal/mol as compared to 9.0 kcal reported earlier. Alcohols dehydrated V-amylose hydrate with apparent occupation of the interior of the helix through interaction with glycosidic oxygens. On the other hand, water, ammonia, and formaldehyde appeared to interact with hydroxyl groups on the exterior of the helix. Further study showed that interaction of V-amylose and ammonia was not completely reversible. Absorption of ammonia was greatest at high pressures and low temperatures. More detailed investigation of selective reactions between V-amylose and small molecules will be carried out under a grant to Arizona State University.

Initial studies under the grant to Purdue University for research on disulfide bond cleavage were directed to developing methods necessary for the evaluation of the effect of alkali on proteins. A procedure for amide nitrogen analysis was developed wherein cystine residues are first reduced with bisulfite to eliminate interference of cystine during alkaline hydrolysis of amides. Conditions for treatment of tryptophan with sodium nitrite were optimized to render the Spies and Chambers colorimetric determination of tryptophan in protein more precise. Proteins of both corn and wheat will be studied under this grant.

Studies on zein protein under a PL 480 grant to the National Institute of Agronomic Research, Paris, France, have been completed. Zein was found to be complex. Although amino acid analyses indicate the presence of only two principal protein components, an array of heterogeneous fractions arises from the association of these proteins in various combinations with relatively small peptide chains and with variable amounts of pigment. The amount and type of the various protein complexes depends on conditions under which protein is isolated. Thus, the resolution of zein into well-defined and separated protein fractions is a difficult task. Mild conditions of purification are required to obtain reproducible experimental

results. Alkali treatment, such as is used in commercial zein preparation, results in a stable protein-pigment complex that owes its stability to the blocking of protein sulfhydryl groups.

At the University of London, London, England, under a PL 480 grant for research on starch- and glycogen-debranching enzymes, examination of the carbohydrase enzymes in sweet corn revealed an apparently new transferase that can transfer α -1,6 linked maltose units from one pentasaccharide to another. A new enzymatic method has been developed for obtaining the degree of branching and other structural analyses of amylopectins and glycogens.

4. Physical studies on corn components. Nuclear magnetic resonance studies have been undertaken to obtain characterizing information on amino acids and vinyl derivatives thereof and to elucidate effects on the chelating properties of ramulosin (a grain fermentation product) induced by halogen substitution. Infrared spectra of lipids isolated from starch fractions indicate that these lipids are free fatty acids.

Studies on use of dimethyl sulfoxide (DMSO) solutions of amylose for film-casting showed that DMSO-complexed films could be stretched to give oriented X-ray fiber diagrams for use in determining crystal structure. When converted to B-type structure, the DMSO-complexed film exhibited remarkably improved flexibility in comparison to films cast from butanol-water. Tensile strengths, however, were comparable (ca. 8,600 lbs./sq. in.). The number of double folds at 16 percent R.H. was used as a measure of brittleness. For films 1.5 mils in thickness the number of double folds observed was 1,000 for amorphous amylose, and 320 and 105 for crystalline "B"-amylose from DMSO and from water, respectively. V-amylose films were brittle at 16 percent R.H.

Under a grant at Princeton University, a number of basic properties of films from HA starch were measured, including modulus-temperature relationship, stress relaxation behavior, glass transition temperature, and glassy and rubbery moduli. These qualities were related to such factors as percentage of amylose in the starch and presence of plasticizers.

Research has been initiated at Iowa State University under a grant for basic investigations on heat, mass, and momentum transport in cereal starches and flours, but significant results have not yet been obtained.

At the "Giuliana Ronzoni" Scientific Institute of Chemistry and Biochemistry, Milan, Italy, IR and NMR investigations on model compounds and on amylose dissolved in DMSO have established an intramolecular hydrogen bond in amylose between specific hydroxyl groups of adjacent glucose residues. Such a bond not only serves to stabilize the helical form but also has an important bearing on the reactivity of amylose. This work is being conducted under a PL 480 grant.

In studies under a PL 480 grant to the University of Osaka Prefecture, Sakai, Japan, scientists are investigating possibilities of polarography of carbonyl groups as a means of analysis of starch and derivatives. They have succeeded in converting glucose to a polarographically active form, but the method of derivitization is not applicable to starches.

5. Chemical composition of corn grain. Compositional studies on corn are relevant to its industrial utilization. Results are reported under Area 3, subheading A.

B. Chemical and Physical Investigations to Improve Products

1. Reactions of maltose and glucose. Investigation of amide complexing ability revealed that in aprotic solvents isolatable complexes with urea were formed by glucose, maltose, and ribose. The complexes dissociate in aqueous and alcoholic media. N-substituted ureas do not complex. Continuing investigation of this phenomenon showed that in addition to urea, ethanol, dimethyl formamide, dimethyl sulfoxide, 2-oxazolidone, and hexamethyl phosphoric triamide associate with anhydrous maltose to form stable, isolatable molecular complexes. However, only urea formed an isolatable complex with glucose. The α - and β -anomers of D-glucose pentaacetate were found to form a 60:40 constant composition mixture that has a nearly constant melting point and optical rotation. This mixture crystallizes from various solvents even when different ratios of the components are present. It can also be recrystallized from various solvents without changing its properties. A simple method was developed for selective deacetylation (at C-1) of glucose and maltose acetates on silica gel. The structure of a third caramel-flavored compound discovered in studies of model sugar-amine browning reactions has been completely established to be 4-hydroxy-2,5-dimethyl-3(2H)-furanose.

2. Reaction of carbohydrates with mercaptans. Contract research at the University of Arizona on the reaction of mercaptans and carbohydrates has been completed. The most interesting product obtained during the study was the polymer from glucose and 1,10-decanedithiol. Final conclusions are that the best polymers are obtained in hydrogen fluoride (14-50 percent) catalyzed reactions in dioxane at 0-24° C. Dialdehyde starch did not react with the dithiol under these conditions but did yield a highly cross-linked, colored product if the reaction was run in liquid hydrogen fluoride. A variety of modified polymers was obtained by oxidation, acetylation, methylation, and xanthation of the glucose-decanedithiol reaction product. Glucose polythioacetals show some promise as adhesives. A covering patent application has therefore been filed.

3. New derivatives of starch and related carbohydrates. In final phases of research to explore possible methods of synthesizing amino acid derivatives of starch, the reaction of sulfur monochloride with carbohydrates was shown to yield complex mixtures of products containing both chlorine

and sulfur. No successful method was found for separating these mixtures; hence, the reaction is not a practical means for replacing hydroxyl groups of starch or glucose with chlorine. Because halogen derivatives of carbohydrates should be versatile intermediates and might prove to have potential for industrial use in synthesis and for other purposes, a study to find satisfactory methods of preparation of these derivatives has been undertaken.

Research under a grant to Ohio State University showed that 2,3-unsaturated glucose and mannose derivatives could be prepared by elimination reactions based on sulfonic esters (for trans glucose hydroxyls) or thionocarbonates (for cis mannose hydroxyls). Overall yields were 35-50 percent. Thiol derivatives and glucosyl sulfonyl halides could be added to olefinic bonds in sugar molecules. Specific wavelengths of UV light were shown to promote reactions at a sulfur atom in organic molecules without inducing side reactions at oxygen atoms.

Under a second grant to Ohio State University, tetrahydropyran-2-yl derivatives of amylose, amylopectin, and starch were prepared. Also, a series of acetals with various degrees of substitution were prepared from starch and methyl vinyl ether. These acetals were more labile to acid than were the tetrahydropyranyl derivatives. Vinyl ethers were found to exhibit differences in their reactivity toward starch.

At Purdue University, studies under a research grant showed that oxidation of penta-O-acetyl- α -D-glucothiopyranose with sodium metaperiodate yielded a crystalline sulfoxide whereas oxidation with peracetic acid formed a crystalline sulfone.

Contract research at Ohio State University on synthesis of amino derivatives of sugars has been completed. In final phases of the work, 3-amino-3-deoxy amylose was successfully synthesized and its structure was proven by degradation. Also, the corresponding 2-amino derivative was prepared, but time did not permit confirmation of its structure. Basic discoveries made during this work are being exploited under a grant for research on amination of starch with low-cost reagents.

In research under a PL 480 grant to the Plastics Research Institute TNO, Delft, The Netherlands, the preparation of sodium, potassium, lithium, calcium, barium, and magnesium alkoxides of starch by reaction of starch with the corresponding methoxides was studied in detail. Conditions were established for achieving wide ranges of substitution.

Research on fatty acid and fatty amine derivatives of dextrans has been completed by the Institute of Industrial Chemistry, Bologna, Italy. Various surface-active hydrophobic fatty acid esters and hydrophilic fatty amino dextrans were prepared and characterized. Industrial testing in Italy indicated possible uses for the esters as additives to printing inks, as agents for waterproofing paper, and as stabilizers for emulsions. The

amino dextrans promoted adhesion of active compounds to plant surfaces in agricultural spray emulsions. This research was conducted under a PL 480 grant.

4. Graft copolymers. Basic studies showed that the degree of swelling of starch granules strongly influenced the composition of the product obtained in ceric-ion-catalyzed grafting of acrylonitrile to starch. Variation of this parameter provides a further means for modifying properties of graft copolymers. Indeed, for at least one type of starch substrate, a satisfactory reaction requires avoidance of gelatinization.

Saponified graft copolymers of starch and methyl acrylate or acrylonitrile were found to have very high viscosities in solution. Both types of hydrolyzed graft copolymers showed good tolerance to salts. Cupric chloride was found to give the most favorable results when used as a chain modifier in the ceric-ion-catalyzed grafting of acrylonitrile to starch.

In contract research at Stanford Research Institute, studies to determine optimum conditions for grafting the following monomers to starch were completed: vinyl chloride; styrene; methyl methacrylate; vinyl acetate; and methyl, ethyl, and butyl acrylates. Good add-on and minimum formation of homopolymer were achieved for all except vinyl acetate and chloride, which gave no significant grafting under any conditions tested. Rate constants for the decay of free radicals were determined and activation energies for grafting were evaluated. Rate equations were developed for grafting of several monomers. (Pilot-plant studies on graft copolymers are reported under subheading 1-D-3.)

Under a PL 480 grant to the Hebrew University, Jerusalem, Israel, scientists have investigated the effects of reaction variables on length and frequency of grafts, monomer add-on, and product yield in the anionic grafting of ethylene and propylene oxides, acrylonitrile, methacrylonitrile, and methyl-methacrylate to starch and dextrin. The influence of composition and structure of the graft copolymers on properties such as solubility, viscosity, and polymer melt temperature were established. Some of the products have properties indicative of utility in one or more industrial applications. However, cost of the products, which will be relatively high owing to the use of anhydrous DMSO as solvent for the reactions, must be determined before their commercial potentialities can be appraised.

5. Thermal reactions of starch. The problem of heat transfer at low pressure in the fluidized bed reactor for converting starch to levoglucosan was successfully solved. The reactor can now be operated in stable equilibrium conditions for unlimited times. An outgrowth of this work is the possibility of activating starch for reaction by utilizing its dielectric properties. A grant to investigate this possibility has been made to the University of Pittsburgh.

In studies under a PL 480 grant to the University of Edinburgh, Edinburgh, Scotland, the thermal stability of starch and its components from 150° to 350° C. was investigated in order to characterize various types of starches and their components. In the presence of small amounts (2 percent) of simple inorganic salts such as sodium chloride and bicarbonate, the threshold temperature of decomposition was significantly lowered and production of volatile products was increased.

6. Starch polyol foams. Studies on rigid urethane foams based on starch polyols showed that stability to accelerated aging (4 weeks at 70° C. and 100 percent relative humidity plus 1 week at 100° in a forced-air oven) equaled that of commercial foams. Addition of a commercial organophosphorus compound to the polyether system imparted fire retardancy to the final foam. Rigid foams containing over 90 percent open cells were obtained by use of α -sulfostearic acid for neutralization of the alkaline catalyst employed in preparing polyols by the reaction of propylene oxide with glycol glycosides. It was also discovered that foams with acceptable stability can be made when up to 35 percent of a commercial gum-type corn starch dextrin is included in the formulation. These foams are self-extinguishing without addition of conventional fire retardants. (Pilot-plant studies on starch polyol foams are reported under subheading 1-D-2.)

7. Chemical products from starch. In experiments with cyanoethylated pearl starch, partial hydrolysis to acid and/or amide groups and limited alkaline hypochlorite oxidation of starch hydroxyls to carbonyl and acid groups gave a series of derivatives with viscosity characteristics suitable for paper-coating applications. Permanganate oxidation of starch eliminated set-back tendencies of pastes but, the oxidized starches were unsuitable as paper-coating adhesives because of poor paste clarity and deposition of sediment on cooking.

Up to 36 percent of the relatively high-priced isolated soy protein could be replaced with the less expensive soy flour if a small amount of dialdehyde starch (DAS) was included in a conventional formulation for pigmented paper coatings. Very good wet-rub resistance was obtained and other coating properties such as brightness and wax pick resistance were essentially unaffected. Exterior exposure studies showed good durability (7 months) of southern pine plywood bonded with protein-DAS glue. All specimens failed in 9 months, whereas commercial resin-bonded plywood remained in good condition for 11 months and showed failure of one-third of the specimens during the 12th month.

Products obtained by crosslinking starch xanthate in water with high-molecular-weight polyethylenimine were incorporated with pulp into paper handsheets. Improvement in wet- and dry-strength was comparable to that achieved with starch xanthide. Retention was 100 percent. Several starch-based products were successfully used as reinforcing agents for natural and synthetic rubbers. More detailed evaluation will be performed by the University of Akron under a research contract.

Contract research on retention of starch xanthates and xanthides by wood pulp was initiated at the Institute of Paper Chemistry. First results revealed that starch xanthate of 0.12 D.S. was not significantly retained on bleached softwood kraft fibers under a wide range of fiber concentrations, pH, alum content, and contact time.

At the Ahmedabad Textile Industry's Research Association, Ahmedabad, India, various hydroxy ethyl glucose derivatives are being prepared for use as reference compounds in studies designed to elucidate the structure of hydroxyethylated corn starch. In other research at this institution, a practical procedure was devised for preparing copolymerization products of starch and locust bean gum and of starch and guar gum. These investigations were conducted under PL 480 grants.

8. High-amylose (HA) starch films and chemical derivatives. A commercially feasible procedure was developed for preparing the triacetate of HA starch. Hydroxypropyl, methyl, and carboxymethyl ethers of HA starch were prepared for evaluation of film properties. Activation of HA starch by pretreatment with dimethyl sulfoxide was the most effective method for achieving rapid acetylation or chlorination with phosphorus pentachloride. The chlorinated starch could be converted to starch amines by reaction with aliphatic amines.

C. Microbiology and Fermentation

1. ARS Culture Collection. As of January 1, 1966, the ARS Culture Collection contained 17,070 permanent cultures. During 1965, a total of 2,228 cultures were distributed to domestic and foreign recipients. Twenty-four samples of reportedly toxic fescue were examined for unusual fungi not common to nontoxic fescue or to orchard grass. Approximately 70 isolates were retained. (See Area 17 of this report.) All new type-cultures (88) of Aspergillus described in the new book on this genus by Raper and Fennell (University of Wisconsin) have been deposited in the culture collection.

Contract research by the American Type Culture Collection revealed that 94 percent of the fungi scheduled for study remained viable after freezing and storage for 6 months in liquid nitrogen. Those that failed this test did survive when 10 percent dimethyl sulfoxide was substituted for 10 percent glycerol as the suspending medium.

Research of importance to the ARS Culture Collection is in progress at several foreign institutions under PL 480 grants. At the National Institute of Agronomic Research, Madrid, Spain, studies have been initiated on the distribution of aerobic actinomycetes. Techniques have been developed for the isolation, characterization, and assay for antibiotic-producing activity of soil isolates. The taxonomic characteristics of 49 soil isolates of actinomycetes, including electron photomicrographs of spores of representative cultures, were submitted to the Northern Division. The cultures themselves were likewise submitted for further study.

Aerobic actinomycetes are being collected by scientists of the Central Drug Research Institute, Lucknow, India, with particular emphasis on strains producing antibiotics active against Agrobacterium tumefaciens, the organism responsible for galls on many economic plants. About one-third of the actinomycete cultures isolated so far inhibited the growth of A. tumefaciens on agar media. These cultures represent various soil types. Initial experiments to produce the antibiotic in liquid fermentation media gave variable results.

At the University of Allahabad, Allahabad, India, additional new species of the order Mucorales were discovered. In other studies at the University of Allahabad, it was established that there is no significant difference in yields of fumaric acid, itaconic acid, penicillin, and amylase from lyophilized and nonlyophilized cultures of the respective fungi producing those materials. There has been no marked reduction in viability of lyophilized cells of selected species of Streptomyces during storage for 1 year.

2. Taxonomic investigations. In taxonomic studies of Mucorales, the zygosporic stage of the species Rhizopus chinensis has been discovered for the first time. This and other small-spored species of the R. chinensis group have now been found in molded peanut meal. During the work, a disease of Rhizopus was observed in which Rhizopus is attacked by species of the genus Syncephalis. Some species are strongly parasitic on R. oligosporus, the tempeh mold. A method for growing isolates of Syncephalis in pure culture was discovered. Electron microscopic studies showed that no Rhizopus strain examined had sporangiospores with smooth walls. This observation casts doubt on the validity of the "smooth-spored" criterion in taxonomy of these microorganisms.

In studies on yeasts, characterization of the protosexuality of Hansenula wabatongushiensis has been completed. Hybridization was found to be the most objective of all criteria for taxonomic speciation of yeasts.

3. Studies on enzymes. Studies made of the relative proportions of the two glucoamylase isozymes produced by Aspergillus niger NRRL 3112 showed that one predominates in the culture for up to about 48 hours. Later, the second becomes predominant and continues to increase for up to 96 hours. After the first 48 hours there seems to be no further increase in the amount of the first isozyme; in fact, drastic decline is occasionally observed. Glucoamylases from different strains of black Aspergilli were found to be similar. A method was developed for automatic assay of glucoamylase.

Research under the grant to the University of Nebraska showed that Rhizopus delemar produces carbohydrase isozymes as does Aspergillus niger. All glucoamylases from A. niger had similar molecular weights ($125,000 \pm 6\%$) as determined by ultracentrifugation but isoelectric points were different.

These isozymes also had identical action patterns and N-terminal amino acid residues (L-alanine) and were apparently stable to reagents that promote dissociation into subunits.

Research on coupled enzyme systems was initiated with studies on formation and properties of a glucose isomerase produced by Aerobacter aerogenes and a stable D-glucose 6-phosphate dehydrogenase produced by Neisseria perflava. The glucose isomerase appears to be phosphohexoisomerase. The glucose-6-phosphate dehydrogenase was substantially purified but separation from myokinase activity was not achieved. At all stages of purification, relationship of dehydrogenase activity to nicotinamide-adenine dinucleotide (NAD) or to nicotinamide-adenine dinucleotide monophosphate (NADP) as coenzymes remained constant. This enzyme, which is the first NAD/NADP-dependent glucose-6-phosphate dehydrogenase discovered in a gram negative organism, possesses remarkable stability that should greatly simplify its evaluation for converting cereal grains to industrial materials.

In other studies to find enzymes that might be useful for chemical conversions, a mutant strain of Candida tropicalis Y-1367 was found capable of producing 9,000 units/ml. of glucosyl transferase. This enzyme, which converts glycerol in solutions containing dextrin to glycerol glucoside, was purified 100-fold, making possible more accurate determination of several of its characteristics. Several promising strains of organisms capable of producing D-glucarate dehydratase have been isolated from soil.

Initial studies on spores as agents for effecting chemical conversions involved a survey of 20 species of molds. Spores from all species showed metabolic activity on most of the substrates tested, including fatty acids, other organic acids, amino acids, hydrocarbons, and terpenoids. With each of several preparations tested, spore activity increased with age. These results are encouraging not only because activity has been demonstrated but also because a wide variety of substrates can be metabolized by spores.

Under a grant to Iowa State University, action patterns of amylases from a number of microorganisms are being determined. Amylase from Thermoactinomyces vulgaris converted amylose primarily to maltotetraose. That from Streptococcus bovis converted starch first to maltotetraose and maltose; later, significant amounts of maltotriose and glucose were formed.

At Kansas State University, also under a grant, a new method for staining acrylamide gels was developed that will accelerate analysis of enzymes obtained by preparative gel electrophoresis.

The program of research on enzymes has been broadened by a grant to the University of Arkansas for basic studies on the mechanism of enzymatic hydrolysis of cereal starches. Research under this grant has not yet been initiated.

Lytic enzymes of microbial origin are being studied under a PL 480 grant to the University of Liege, Liege, Belgium. Three distinct lytic endopeptidases from the Streptomyces albus G enzyme complex have been isolated and characterized. They are highly active on lysine-containing cell walls of gram positive bacteria. By use of these specific endopeptidases in conjunction with other enzymes from the Streptomyces complex, structures of four different types of peptide bridges linking peptidoglycan units of cell walls were elucidated.

Work on the mechanism of action of the $F_1 \beta$ -1,4 endo-N-acetylmuramidase from Streptomyces showed that (1) pH optimum for lytic activity depends on the structure and composition of the bacterial wall, (2) affinity of the F_1 enzyme for N-acetylmuramyl linkages is greatly enhanced by peptide substitution of these residues, (3) few walls from gram positive bacteria possess the tight network peptidoglycans found in Staphylococcus aureus and Micrococcus roseus.

4. Biological insecticides for Japanese beetle. Refractile spores of Bacillus popilliae have been repeatedly produced in liquid media containing 1 percent of activated carbon. Yields range from 10,000 to 100,000 per ml. These spores have withstood drying in sterile soil and storage for 6 months. They have also survived for at least 4 months in sterile sand and in a $CaCO_3$ -talc mixture like that used in commercial spore preparations. The most consistent sporulation in shaken cultures was achieved with activated carbon made from domestic nut shells; different activated carbons differed greatly in effectiveness.

A new, infective derivative strain, B-2309M, sporulates consistently on agar medium. About 10-20 percent of the cells on a plate form spores.

Improvements in media have resulted in more vigorous and reproducible growth of B. popilliae. Populations of 1.6 billion viable cells per ml. are now regularly attained in 18-20 hours.

Study of sporogenesis of B. popilliae in larvae showed exclusive and limited vegetative development in the hemolymph for the first 4 or 5 days. At this time, pre-spore forms abruptly appear and predominate, followed by sporulation to about 80 percent by the ninth day. Sporulation finally reaches 90-95 percent by subsequent slow accumulation from vegetative cells remaining after the initial sporulation.

Investigation of the proteins of grub hemolymph revealed a high-molecular-weight major fraction plus small amounts of four other protein fractions of lower molecular weight. The latter increase in concentration at the expense of the high-molecular-weight fraction during the course of milky disease. Also, the oxygen level in hemolymph was found to increase during sporulation of vegetative cells.

Serological studies based on successful preparation of rabbit antisera to surface antigens of vegetative cells and spores of B. popilliae and B. lentimorbus have been undertaken. The most significant of the initial results are that both spores and vegetative cells possess a common antigen; that antigen content changes with cell age; and that B. popilliae vegetative cells are not antigens for spore antiserum. These results may provide a way to detect the point at which commitment to sporulation occurs.

In the contract research at Baylor University, electron microscopy of thin sections of cells of B. popilliae in different stages of sporulation, together with phase microscopy of sporulation on solid media, is providing a highly detailed picture of the entire process and of the intimate structural changes that transpire. Spores prepared by the NU plate culture procedure were shown to be true spores. Significant structural differences between vegetative cells from infected larvae (pre-symptom stage) and those grown in the laboratory were observed.

Study of enzymes in sporulation under the contract at Michigan State University involved a comparison of enzymes of spore-like bodies produced by B. popilliae and B. lentimorbus cultured on media containing barbiturate or β -hydroxybutyrate (BHB). Extracts of spores produced in vivo and of spore-like bodies produced in vitro had similar enzyme activity. Activity was found generally to be influenced by BHB, although changes in activity for corresponding enzymes from the two parent microorganisms were not parallel. As much as 80-90 percent "sporulation" of B. popilliae was obtained in a liquid medium containing BHB. The "spores", however, could not be germinated. Barbituric acid was found to specifically influence transition of vegetative cells in liquid media to spore-like bodies.

Contract research at Kansas State University has resulted in a lyophilization procedure that achieves survival of up to 83 percent of vegetative cells of B. popilliae.

5. Microbial polysaccharides. Engineering research on continuous fermentation with B-1459 showed that polymer formation is not associated with cell growth but takes place during the entire fermentation at an essentially constant rate. A pilot-plant two-stage continuous fermentation was operated for 5 days with an overall yield of 62 percent based on glucose added. Microbial contamination was present, but did not appear to affect the viscosity of the product in comparison to uncontaminated continuous runs. In a 6-day run, modified by intermittent addition of inoculum, polymer yield was about 60 percent. In this experiment, microbial contamination occurred at 2 days but did not influence the fermentation until the fifth day. Based on kinetic studies on the growth of the B-1459 organism, an improved batch fermentation procedure was developed that reduces fermentation time from 96 to 48 hours.

Under the grant to Cornell University, electro-deposition was used successfully for removal of polysaccharide B-1459 from fermentation broths. Rates

of 0.5 mg/cm²/hr could be obtained. It was also observed that resting cells produced polysaccharide in the same yield as growing cultures. Toxicity of hydrocarbons below C₁₁ for use in emulsion-type fermentations was confirmed.

Studies on the structure and chemistry of polysaccharides showed that the β -D-mannuronic acid linkage, found in B-1973 and certain other polysaccharides, possesses inherent instability that can lead to anomalous chemical behavior. Although the mode of linkage of pyruvic acid appears to be the same in polysaccharides B-1828 and B-1459, the percentage of this acid in B-1828 is more consistent among various preparations than in B-1459 and the stability to acid hydrolysis is much greater. Structure studies on Y-6493 and Y-6502 phosphogalactans have been completed. Results show that phosphate occurs exclusively as α -D-galactosyl-1-phospho-6'-galactosyl end groups. The remaining galactosyl linkages are α -1,3 and α -1,6 in about equal proportions. A chemical-chromatographic technique has been developed that simplifies identification and quantitation of complex mixtures of monosaccharides obtained by acid hydrolysis of polysaccharides. The aldoses are reduced to alditols, fully acetylated and separated by gas-liquid chromatography.

The extracellular polymer produced by Y-6272 has been separated into three components: a galactomannan, a glucomannan, and an N-acetyl glucosamine polymer that is combined or complexed with amino acids. Optimal conditions were established for production of this polymer.

Structure studies were strengthened by a grant to the University of Wisconsin for studies on the fine structure of polysaccharide B-1459.

It is timely to note that the Northern Division's research on microbial polysaccharides appears to have founded a new type of industry. Evidence has been accumulating which shows that industrial activity has transcended interest merely in the Northern Division's specific polysaccharides. Stimulated by our research and development program in this area and, undoubtedly, by the favorable acceptance of B-1459 as a commercial product, industry has gone on to independent research and promotion of independently discovered products. For example, one company is actively investigating derivatives and modifications of B-1459. Several companies are screening new microbial sources. Two others are actively seeking markets for products which they themselves discovered and developed. If this trend continues, microbial polymers may join antibiotics as industries opened up by the Northern Division's research.

6. Plant antibiotics. Preliminary results of tests conducted by the Boyce Thompson Institute on plant antibiotic preparations furnished by the Northern Division showed that several were effective against one or more of 12 economically important fungal diseases of plants. One (F-17) inhibited six diseases, but none was active against either Dutch elm disease or nematode infestation.

7. Microbiological processes and products. In studies on genetic control of fermentation, a phenomenon of spontaneous reversion from the mutant state to "wild-type" at high incidence upon dilution of cultures was discovered. Unrecognized, this new phenomenon appears to have misled previous investigators who had reported conjugation in Pseudomonas aeruginosa. The phenomenon was found using the mating-type strains and the system on which reports of conjugation in the literature were based, as well as with new mutant strains prepared at the Northern Division. It is concluded that conjugation in P. aeruginosa has not been established. However, the pronounced effect of dilution on reversion is unique and may offer a clue to the genetic processes of pseudomonads.

The strain of Hansenula holstii NRRL Y-2448 used for the production of phosphomannan is diploid. By appropriate techniques, haploid strains now have been derived from it which produce either little or much phosphomannan. When these haploids are judiciously crossed, new diploid cultures are obtained which produce phosphomannans which differ qualitatively and quantitatively from that of the parent cultures. Results of genetic analysis suggest that there are several lethal factors which affect spore formation and germination. Selective breeding experiments are being done to overcome this restriction in spore viability without changing the inherent capacity of the organism for polymer production. If successful, more complete analysis of factors affecting phosphomannan formation can be done and perhaps modifications of phosphomannan capacity can be introduced by breeding and by mutation.

In the Pioneering Laboratory for Microbiological Chemistry, investigation of a genetic defect of Rhodospirillum rubrum that interferes with identification of mutants on the basis of colonial morphology revealed that the lipopolysaccharide content of cell walls may be implicated in this phenomenon. Preliminary studies of an ornithine-containing lipid present in large quantities in R. rubrum showed that it has no free carboxyl and only one free amino group. Although it behaves chromatographically like a phospholipid, it does not contain phosphorus. Experiments with radioactive tracers suggest that this lipid is related to the arginine biosynthetic pathway. Further investigation of macromolecules responsible for agglutination of certain yeast strains confirmed that disulfide bonds have a specific function in the sex-specific agglutinin and may, in fact, be involved in the agglutinative site.

In other work in the Pioneering Laboratory, a new acid of m.p. 130° was isolated from the extracellular lipid produced by the yeast YB-2501, the taxonomic position of which has not yet been established. The acid was shown to be 13-keto-8,9-dihydroxydocosanoic acid by periodic acid cleavage to two aldehydes which on oxidation yield suberic acid and 5-ketotetradecanoic acid. The configuration of the 8,9-hydroxyls was shown to be erythro by conversion of erythro-8,9,13-trihydroxydocosanoic acid of m.p. 156° to the new acid via oxidation of the 13-hydroxyl group after acetonation of the 8,9-hydroxyls. A detailed study of the hydrogen

bonding system in ramulosin is in progress. Eleven halogen derivatives have been prepared and subjected to ultraviolet, infrared, and NMR analysis. Infrared and NMR examination of ramulosin itself indicated that it exists partly as the 6-membered hydrogen chelate and partly as a 12-membered dimer.

In studies under a PL 480 grant at the University of Milan, Milan, Italy, the levels of 2-ketogenic and of 5-ketogenic activities in Acetobacter suboxydans were found to be correlated to particular vegetative states, rather than directly determined by external inducers such as substrates (glucose, gluconate, sorbitol, mannose, glycerol), amino acids, or vitamins. It was concluded that these enzymatic activities are constitutive in the organism. Therefore, it does not appear that fermentation conditions can be established by which only 5-ketogluconic acid will be produced. This research has been completed.

At the University of Newcastle upon Tyne (formerly University of Durham), Newcastle upon Tyne, England, scientists have purified the teichoic acid of Streptomyces griseus and shown it to be a mixed polyphosphodiester of glycerol and ribitol. The gram positive bacteria, in contrast, contain separate polyglycerol and polyribitol phosphates with alanine ester and glycosyl groups linked to the polyols. The S. griseus teichoic acid contains β -glucopyranosyl substituents on some of the ribitol units and small amounts of acetic and succinic monoesters in place of alanine. Alkaline hydrolysis and phosphomonoesterase treatment gave rise to two new phosphate esters: ribitol 3-phosphate, which is resistant to hydrolysis by alkaline phosphomonoesterase; and ribitol 3-phospho-1'(3')-glycerol. This research is being conducted under a PL 480 grant.

8. Fermentation acids. Research on fermentation acids is in progress under PL 480 grants to several foreign institutions.

At the University of Lodz, Lodz, Poland, mutation of Aspergillus terreus by radiant energy yielded several strains of the organism with improved ability to produce itaconic acid. Yields of itatartaric acid per se in the fermentation are still rather low, but this may be due to instability of the acid. Indoleacetic acid was found to stimulate both itaconic and itatartaric acid.

In research at the University of Tokyo, Tokyo, Japan, approximately 1,000 cultures of yeasts, molds, streptomycetes, and bacteria were screened for production of mevalonic acid. Nearly all of the organisms formed some mevalonic acid; however, only members of the genera Aspergillus, Hansenula, Pichia, and Endomycopsis produced significant quantities. Although no organism produced enough mevalonic acid to provide a basis for economic production, the results do indicate that continued survey, improved cultural conditions, and use of mutant strains may eventually lead to a commercially useful process.

In other research at the University of Tokyo, many strains of bacteria, yeasts, and molds have been screened for their ability to produce tartaric acid. Only two strains of bacteria showed possible production of tartaric acid in small quantities. There is now some question whether or not the acid produced by these organisms is really tartaric acid. If not, a new approach to the problem will undoubtedly be needed.

D. Technology - Process and Product Development

1. Cereal xanthides and xanthates. Studies on stability of the xanthide group in solution showed that when treated with base, model compounds (glycol derivatives) containing a xanthide group and a free hydroxyl group underwent fragmentation to yield thionocarbonates (1,2- and 1,3-structures) or S-alkyl xanthate esters (1,4-, 1,5-, and 1,6-structures). The 1,2-glycol monoxanthide yielded the thionocarbonate on standing without treatment. These studies were extended to amylose xanthate and to an amylose-derived model compound blocked in the 6 position. Both compounds rearranged in a manner similar to that of simpler models to yield the thionocarbonates. However, with amylose xanthate itself, which contained no blocking group, the ring of the initially formed thionocarbonate opened with further reaction to yield intermolecular, acyclic thionocarbonates. A byproduct of these studies was development of a facile method for preparing cyclic thionocarbonates that are valuable intermediates for introduction of unsaturation into carbohydrates. In other studies on stability, it was observed that treatment of a precipitated xanthide with dilute acid or a cation exchange resin followed by drying at 80° C. gave products that did not change during storage for several weeks. When starch xanthide (D.S. 0.12) was suspended in water and drum dried at 110-115° C. for 2 minutes, there was no loss of sulfur. The dried products contained 6-8 percent moisture and did not lose sulfur during storage for 4 months at room temperature.

A continuous process has been developed in which starch xanthate is cross-linked ex situ with chlorine gas and immediately metered into wood pulp flow lines. Results with this process were superior to those obtained with the continuous in situ process described in last year's report.

In cooperation with Forest Products Laboratory, 500 lbs. of unbleached southern sulfate pulp was refined according to the Northern Division's specifications. This pulp, which was typical of linerboard stock, was used in evaluation of the continuous process for incorporating xanthide. Results indicating definite potential for promoting crush resistance were obtained in tests at 90 percent R.H. of linerboards containing starch xanthides crosslinked by all of the principal processes developed at the Northern Division. Xanthated commercial corn flour was nearly equal to starch xanthate in improving linerboard.

A large pilot-scale papermaking machine and related equipment have been installed at the Northern Division. With this facility xanthates, xanthides, and other products made from cereal starches and flours can be evaluated under conditions closely resembling those existing in commercial plants of the paper industry.

Contract research at Battelle Memorial Institute on pilot-plant production of xanthides has been completed. Final phases of the work showed equal effectiveness and perhaps a slight cost advantage for chlorine as compared to hypochlorite for crosslinking. Hydrogen peroxide had no functional or economic advantages over chlorine and hypochlorite. Several methods of preparing ex situ crosslinked xanthides were devised. Although the ex situ preparations displayed activity as wet-strength agents in paper equal to that of in situ preparations, they are too expensive and have inadequate shelf life to be commercially acceptable.

2. Starch polyol foams. Contract research on scaling up the starch polyol process, preparing rigid urethane foams with continuous foam-making machines, and evaluating the products has been successfully completed by the Archer Daniels Midland Company. Batches as large as 1,000 pounds were prepared. The foams had good dimensional stability and flame resistance. Their properties equaled those of foams based on methyl glucoside and were superior to those of foams based on sorbitol. Preliminary design and plant investment and operating cost calculations were completed by Archer Daniels Midland Company. Plant gate cost of the polyol for a plant producing 10,000,000 pounds per year was estimated at about 15 cents per pound. (See also subheading 1-B-6.)

3. Graft copolymers. Under a research contract, two 100-pound lots of starch-acrylonitrile graft copolymers were prepared at Stanford Research Institute in its continuous reactor. Irradiation with gamma-rays was found to be less efficient than irradiation with an electron beam in the large-scale preparation of starch-acrylamide graft copolymers. Construction of a pilot plant for continuous production of this copolymer by electron beam irradiation was completed. On the basis of a study of the effects of reaction variables, two 100-pound lots of each of two different starch-acrylamide copolymers were prepared in this pilot plant. (See also subheading 1-B-4.)

4. Starch derivatives for protective coatings. Contract research at Battelle Memorial Institute has shown that four starch derivatives--cyanoethylated corn starch, potassium carboxymethyl starch, a commercial cationic starch, and a commercial hydroxyethylated starch--have promising properties as viscosity control agents in emulsion paints. Extensive evaluation is planned.

Research to find outlets for starch derivatives in the protective coatings industry was strengthened by a contract with Archer Daniels Midland Company.

This contract provides for an investigation of starch-derived glycol and glycerol glycosides in alkyd resins and other coating compositions.

5. Cyanoethylated starch. Large samples of cyanoethylated starch (200 pounds) and cyanoethylated acid-modified starch (450 pounds) were prepared in the pilot plant for use in contract research at Western Michigan University. A simple laboratory process for cyanoethylating starch was successfully devised. In the process, a cake prepared from starch wetted with dilute NaOH solution and dried to 18 percent moisture is allowed to stand in an atmosphere of acrylonitrile vapor. A D.S. of 0.07 was achieved in 50 hours.

At Western Michigan University all experiments in coating and sizing paper with cyanoethylated corn starches have been completed. Samples of all products have been received at the Northern Division, but the contractor has not yet forwarded test results.

6. Corn dry milling. Investigations on dry milling of corn are directed to development of improved products for food and industrial use. Results of this work are reported under Area 2, subheading C-1.

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WHEAT UTILIZATION - FOOD
Northern Utilization Research and Development Division, ARS

Problem. The dominant factor in the wheat economy of the United States continues to be a production capacity that can outpace consumption, including the substantially expanded foreign markets of recent years. Increased exports of wheat from the United States in the last 3 years have brought our wheat carryover to a level that provides less than a prudent reserve. However, the capacity to produce wheat in this country is still restricted.

We view this North American surplus capacity as an unparalleled opportunity. Wheat in excess of domestic needs can be used to buy time in the overpopulated areas of the world until a self-sufficient agriculture can be developed there. Export donations and concessional sales of 600-800 million bushels per year are providing food where it is most needed in the world. This distribution of wheat serves immediate Defense and State Department missions, and also stimulates a long-range market development for United States agriculture. New wheat foods specifically adapted to conditions of use in every region of the world would help materially to popularize this valuable food grain in areas where it is now virtually unknown, and development of simplified methods to process the products at the point of use would speed their adoption.

We also need to increase the commercial exports (currently less than 200 million bushels annually) that contribute favorably to our international trade balance. New processes to elicit maximum quality performance of wheats and flours in products produced in Europe and Japan would help significantly to promote trade in these dollar markets. Sustained further gains in wheat markets are necessary to ease governmental restrictions on production more than they have already been eased, and especially to strengthen export trade balances. Increased world supplies of wheat and restrictive political decisions in the European Economic Community have contributed to seriously reduced commercial exports in some years. Everything possible must be done to increase total wheat markets, but especially those in which payments are made in dollars.

Consumers of wheat foods in this country have benefited greatly by introduction of a wide variety of new and improved products. Well-balanced diets, reasonable food costs, and improved convenience result from such developments and are suitable objectives of research. Domestic per capita consumption has become stabilized over the past 3 years. Research programs along these lines thus sustain and increase markets for wheat.

An essential foundation for a successful product and process development program is basic research on the composition of all classes of wheat and the fundamental properties of their constituents. This kind of information provides the foundation for improved and new products and processes.

USDA AND COOPERATIVE PROGRAMS

The Department conducts a continuing, long-range program of research involving analytical, organic, and physical chemists, biochemists, microbiologists, systematic biologists, and chemical engineers engaged in basic, applied, and developmental studies pertinent to utilization of wheat in food.

The Federal scientific effort (Northern region) for research on food utilization of wheat totals 11.2 scientist man-years. Of this number, 4.7 are devoted to chemical composition and physical properties; 4.7 to microbiology and toxicology; and 1.8 to technology - process and product development.

Research at Peoria, Illinois, on chemical composition and physical properties (3.8 scientist man-years) includes studies on separation and physical and chemical characterization of wheat proteins and on the microscopic and ultrastructure of wheat grains and flours and the effects of various treatments. During the reporting period, a project on the effect of conditioning wheat on characteristics of milled fractions was completed and replaced by research on microscopic and ultrastructure.

A research contract (.9 scientist man-year) is in effect with the Nebraska Agricultural Experiment Station, University of Nebraska, Lincoln, Nebraska, for investigations on varietal variations in kernel properties and milling and fractionation characteristics of wheat.

Research at Peoria, Illinois, on microbiology and toxicology (4.7 scientist man-years) is concerned with development of new fermented foods from wheat, with reduction of the microbial population of wheat and wheat flour and with studies on the production of mycotoxins by Aspergillus flavus and other molds. The work also includes a survey of the incidence of aflatoxin in commercial samples of various grains. A research contract in effect with the Agricultural Experiment Station, South Dakota State University, Brookings, South Dakota, provides for a survey of various species of Aspergilli to find and identify those producing toxic metabolites. A portion of this effort (.2 scientist man-year) is allocated to research on food uses of wheat.

Research conducted at Peoria, Illinois, on technology - process and product development (1.8 scientist man-years) comprises investigations on new techniques for milling and fractionation of wheat to obtain improved products for food and other uses and on methods for reducing radioactive contamination of wheat and wheat flour.

The Department also sponsors research conducted by foreign institutions under grants of PL 480 funds. Research on technology - process and product development involves a grant to the Cereal Research Station, Research Association of British Flour-Millers, St. Albans, England, for investigations on quantitative measurement of properties of wheat that change significantly during conditioning (5 years, 1961-1966). This research was completed during the reporting period.

PROGRAM OF STATE EXPERIMENT STATIONS

A total of 22.0 scientist man-years is devoted to this area of research.

PROGRESS -- USDA AND COOPERATIVE PROGRAMS

A. Chemical Composition and Physical Properties

1. Microscopic and ultrastructure of wheat. Electron microscopy showed that lipoprotein at the protein-starch interface or in the wedge protein of wheat endosperm was substantially extracted with acetone, butanol-water, methanol-ethanol or methanol-diethylether. The extraction was more extensive for soft than for hard wheat. Removal of lipoprotein binds protein and starch and destroys dough-forming properties of flour. It was further shown by electron microscopy that wetting the aleurone layer during steeping of wheat led to extensive changes in cellular organization. This result is believed to be indicative of changes in the status of enzyme systems that originate in this layer.
2. Crosses of soft and hard wheats. Contract research at the University of Nebraska showed that softer lines of wheat exhibited poor milling properties but good air classification properties. The opposite was observed for harder lines. There was low correlation between hardness and protein content for identical lines grown in 1961 and 1964. In the protein of wheats characterized by lower protein content, there was some tendency toward increase of the nutritionally limiting amino acids (lysine, methionine, threonine). An improved liquid extraction procedure was developed for fractionation of endosperm protein.
3. Characterization of wheat gluten proteins. Results, reported under Area 4, subheading A-1, are important to understanding the unique functional properties of wheat flour.
4. Proteolysis inhibition by cereal flours and starches. This research was completed last year. The final report, which was received subsequently, provided additional important information about the water-soluble trypsin inhibitor (TI) that was found in the flours of cereal grains. In order of decreasing TI activity, the cereals studied were buckwheat, rye, wheat, barley, corn, oats, and rice. The concentration of TI in buckwheat was about 1/15 that of soybeans, and the activity of rice was about 1/175 of the activity of soybeans. Studies on the isolation of trypsin inhibitor resulted in a procedure for concentrating the activity. Electrophoretic analysis yielded two fractions with antitryptic activity. Both fractions were peptides (or proteins).

Cooking completely destroyed TI in wheat, rice, and millet and strongly reduced TI in oats, but did not affect TI in buckwheat, rye, barley, and corn. Thus, wheat bread had no TI, but some was retained in rye bread. Malted barley retained activity. The incubation of corn at pH 12 destroyed TI.

The mode of action of TI appeared to be similar for all of the cereals and to involve a strong bonding of the inhibitor to its site of action. Studies on the physiological activity of TI concentrates showed that protein digestion in rats was impaired by high levels of inhibitor equivalent to a greater weight of flour than could be eaten by the animal. Time did not permit the investigators to establish whether destruction of inhibitor would be beneficial for animals on a high-cereal diet. Likewise, complete studies on the laxative effect of TI (or accompanying constituents) were not possible. Relative to human consumption of cereal grain products, any effect of trypsin inhibitor seems very unlikely in view of the relatively low levels of inhibitor activity.

B. Microbiology and Toxicology

1. Reduction of viable microorganisms in flour and flour products. Studies on reducing microbial population of wheat flour showed that heating wheat before milling for 6 hours at 60° C. and 13 percent moisture, in combination with use of chlorine in temper and wash waters, yielded a final flour having less than 1 percent of the microbial count of flour from untreated wheat. Functional properties of the flour were not damaged. This process, which avoids problems and disadvantages encountered in direct treatments of flour, is an attractive means for producing flours suitable for use in uncooked frozen or refrigerated foods and for baby foods and other canned products containing flour.

Examination of 1965 crop wheats and flour samples from 22 representative mills in the Montana-North Dakota and Michigan-Indiana study areas showed that psychrophiles constitute the major part of the bacterial flora of wheat and flour, accounting for as much as 94 percent of the bacterial population in wheat and 70 percent in flour.

2. Fermented wheat foods. Studies on wheat tempeh showed that it retains a large part of the nutritive values of whole wheat and is also enriched with niacin and riboflavin as a result of the action of the fermenting organism Rhizopus oligosporus. Losses in making wheat tempeh were about 8 percent as a result of washing, cooking, and draining, whereas losses from fermentation ranged from 3.3 to 27.3 percent depending on length of incubation. On a dry solid basis, recovery is 88.5 to 64.5 percent. Since losses are mainly in carbohydrate and carbohydrate-like materials, the percentage of protein in tempeh is increased over that in the wheat substrate.

Initial research on products resembling sufu (Chinese cheese) revealed that brining of the fermented substrate, besides inhibiting mold growth and imparting a salty taste, serves also to elute protease from the mold mycelium and thus enables the enzyme to act on protein in the substrate. Neutral compounds cannot replace NaCl or other salts for this purpose, a fact

suggesting that protease may be bound to the mycelium surface by a weak ionic linkage. Two molds used in making sufu grew and released proteases into a medium containing wheat flour and NaCl.

3. Aflatoxin investigations. Studies on toxins produced by molds are important to utilization of wheat in food. Results are reported under Area 3, subheading B-2.

C. Technology - Process and Product Development

1. Milling and fractionation. Fine grinding and air classification of a new variety of Gaines soft wheat ("Nugaines") resulted in a protein shift of 70 percent as compared to 77-80 percent for the regular variety. Barley was found to resemble soft red winter wheat in its response to fine grinding and air classification. Methods were developed for preparation of ground whole wheat and SWW wheat bran having particle sizes equivalent to that of conventional HRW wheat flour. These products are suitable for evaluation as additives for paper either with or without chemical modification.

2. Reduction of radioactive contamination of wheat and milled products. Phosphoric acid was shown to be effective for removing strontium-90 from wheat. As finally optimized, the process requires washing the grain for 3 to 4 hours at room temperature in 0.1 to 0.2 percent phosphoric acid solution followed by rinsing, centrifuging, and drying the grain. The grain can then be conventionally tempered and milled. Washing time may be reduced at least 1 hour by employing mechanical mixers. The used phosphoric acid solution may be decontaminated for re-use with little or no loss of phosphoric acid content by passing it through the cation resin Amberlite IR-120. Other acids such as hydrochloric, nitric, or citric may be used with similar results if phosphoric acid is not available. When field-grown wheat with a relatively low initial strontium-90 content (395 pCi/kg) was treated by this method, a reduction of strontium-90 in the wheat of 73 percent was obtained. This was reflected in reductions of 76 percent in bran, 49 percent in shorts, and 21 percent in the clear and patent flours. Use of fine grinding and air classification resulted in further reduction in radioactivity of flour. These studies have progressed as far as is practical with available materials. Any further useful extension of this work would require simulation of highly contaminated wheat samples such as would be anticipated from massive radioactive fallout. This research, which was carried out with the cooperation of the Health and Safety Laboratory of the AEC, has accordingly been terminated.

3. Quantitative measurement of wheat conditioning variables. This research is being conducted under a PL 480 grant to the Research Association of British Flour-Millers, St. Albans, England. Results showed that tempering moisture has a marked effect on endosperm breakdown into particles below

35 μ in diameter and that the response varies with the type of stress applied. Thus, in roller milling, breakdown is greater at high moisture levels; whereas in impact milling, breakdown increases at low endosperm moisture. A subaleurone fraction with an average protein content of 44 percent and making up about 5 percent of the flour was recovered from coarse air-classified flour.

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WHEAT UTILIZATION - FOOD

Western Utilization Research and Development Division, ARS

Problem. The dominant feature of the wheat economy in the United States continues to be a production capacity that can outpace consumption, including the substantially expanded foreign markets of recent years. Increased exports of wheat from the United States in the last three years have brought our wheat carryover to a level that provides less than a prudent reserve. However, the capacity to produce wheat in this country is still restricted.

We view this North American surplus capacity as an unparalleled opportunity. Wheat in excess of domestic needs can be used to buy time in the overpopulated areas of the world until a self-sufficient agriculture can be developed there. Export donations and concessional sales of 600-800 million bushels per year are providing food where it is most needed in the world. This distribution of wheat serves immediate Defense and State Department missions and also stimulates a long-range market development for United States agriculture. New wheat foods specifically adapted to conditions of use in every region of the world would help materially to popularize this valuable food grain in areas where it is now virtually unknown, and development of simplified methods to process the products at the point of use would speed their adoption.

We also need to increase the commercial exports (currently less than 200 million bushels annually) that contribute favorably to our international trade balance. New processes to elicit maximum quality performance of wheats and flours in products produced in Europe and Japan would help significantly to promote trade in these dollar markets. Sustained further gains in wheat markets are necessary to ease governmental restrictions on production more than they have already been eased, and especially to strengthen export trade balances. Increased world supplies of wheat and restrictive political decisions in the European Economic Community have contributed to seriously reduced commercial exports in some years. Everything possible must be done to increase total wheat markets, but especially those in which payments are made in dollars.

Consumers of wheat foods in this country have benefited greatly by introduction of a wide variety of new and improved products. Well balanced diets, reasonable food costs, and improved convenience result from such developments and are suitable objectives of research. Research programs along these lines would sustain and increase markets for wheat.

An essential foundation for a successful product and process development program is basic research on the composition of all classes of wheat and the fundamental properties of their constituents. This kind of information provides the foundation for improved and new products and processes.

USDA AND COOPERATIVE PROGRAM

Research on utilization of wheat for food seeks to solve the most urgent problems hindering the development of markets for the full productive capacity of U.S. agriculture. The emphasis is on (1) expansion of overseas dollar markets for U.S. wheats; (2) development of new wheat food products for long-term market development in food-short nations abroad; (3) raising the domestic consumption of wheat foods by increased variety, quality, and convenience; and (4) finding means to upgrade wheat millfeeds to recover fractions of nutritious food quality. Basic research on the fundamental chemical and physical properties of wheat and barley constituents and on the functional properties of wheat flour constituents supports the product development and problem-solving segments of the program.

Research is conducted by the Western Utilization Research and Development Division at Albany, California; under contracts and grants at Pullman, Washington; Chicago, Illinois; Manhattan, Kansas; Madison, Wisconsin; St. Paul, Minnesota; Menlo Park, California; and Corvallis, Oregon; and under P.L. 480 grants in England, France, Poland, Italy, Australia, Switzerland, Israel, India, Japan, and Belgium.

The Federal program of research in this area totals 27.0 scientist man-years and 8 contracts and grants providing research at a rate of approximately 7.1 scientist man-years per year. Of this number, 7.5 are assigned to investigations on chemical composition and physical properties; 4.7 on flavor; 5.6 on color, texture and other quality characteristics; 0.2 on microbiology and toxicology; and 9.0 on technology - process and product development. In addition, the Division sponsors 13 research grants under Public Law 480.

PROGRAM OF STATE EXPERIMENT STATIONS

A total of 22 scientist man-years is devoted to this area of research.

PROGRESS -- USDA AND COOPERATIVE PROGRAMS

A. Chemical Composition and Physical Properties

1. Proteins of Wheat. We study wheat proteins and components that associate with them because the unique character of wheat protein is essential in making a strong, smooth, resilient dough and baking a bold attractive loaf of bread. The spread of cookies, the formation of cake crumb, and the texture of crackers also depend considerably on the amount and character of wheat protein, although each demands differences in these factors from those ideal for bread making.

In former days when baking was a hand operation, the baker adjusted his formula and dough manipulation to correct for differences in the quality of the flour he used. If a dough was sticky, the baker would just add a little more flour; if it was a little tough he would add more liquid or shortening

or increase the mixing time. The need then to control baking quality of flour was not as great as it is now. In large-scale mechanized bakeries with mechanical continuous dough mixing, the skilled hands of a baker are no longer available. Uniformity of raw material is the only way to provide efficiency in the baking operation. This is the reason we undertake basic studies of the constituents of wheat and flour and the relationship of constituents to the baking quality.

Of the individual constituents of wheat flour, the proteins have been, and still are, receiving the greatest attention from cereal chemists. Aside from the nutritional importance of flour protein, it is known that the physical and structural characteristics of doughs, batters and all baked products made therefrom are, in large measure, determined by the nature of flour proteins. It is not yet known, however, how the proteins exert their effects.

We study wheat proteins by investigating differences in their composition, physical properties, and chemical reactivities. Individual proteins are separated by differences in solubility, size, and electrical charge by means of solvent differentiation, column chromatography, and electrophoresis. They are characterized by composition, specific immune reactions (antigenicity), and specific enzymic properties.

We have compared soluble and gliadin proteins from hard red spring and durum flours, flours which differ markedly in baking performance and in protein content. From these dissimilar flours, all antigenic proteins of similar migration rate under electrophoresis showed immunochemical cross-reaction, whereas proteins of dissimilar migration rates did not. From this we conclude that individual antigenic proteins of similar migration rate in the two wheat types must be similar in composition and structure. All this means is that the soluble and gliadin proteins are the same in all wheats. Differences in baking performance must, then (1) be related to amounts of protein components rather than type of protein, (2) be influenced strongly by non-protein components, or (3) reside completely in the insoluble glutenin protein fraction.

More accurate quantitative estimates of the distribution of various proteins amongst flours now appear to be a major goal for research. It is possible, however, that non-antigenic portions of the protein molecules may cause differences in properties. Of immediate need for attention are further studies of enzymic activities of separated proteins.

Dr. Grabar, working under a P.L. 480 grant at the Pasteur Institute in Paris, is using the immunochemical methods of analysis that he pioneered to compare proteins in various cereals. He has shown that several of the proteins in barley, rye, and wheat are identical in their immunochemical reactions. Proteins in maize and oats were formerly thought to be in the same group as the barley, rye and wheat proteins, but they are only partially similar. He has shown that one group of proteins in malt is derived from a similar

group existing in the barley prior to malting, even though the changes that occur during malting tend to obscure this fact when proteins are characterized by other methods. He has shown that the parts of certain protein molecules that are responsible for antigenic specificity are not the same parts of the molecule that are responsible for specificity of enzymic activity.

At the University of Poznan in Poland, a P.L. 480 research grant project on the dependence of wheat enzyme activity on sulfhydryl groups has been concluded. The relationship between sulfhydryl groups and enzyme activity is well known, but details of this relationship in cereal enzymes had not been previously reported. In this study, protein-splitting enzymes of wheat flour were isolated and characterized, particularly in regard to their sulfhydryl and disulfide contents. Observations were made on the stability of sulfhydryl groups in whole and ground wheats and malted wheats during storage, on changes in sulfhydryl content during soaking prior to germination, and on comparative effectiveness of various sulfhydryl blocking reagents added to flour or ground whole grain suspensions. A purified enzyme, protease A, was studied. Its sulfhydryl groups were detectable only in the presence of ammonia, a finding that may lead to a chemical characterization of just how the sulfhydryl groups may be masked in proteins, where we know their presence is essential to enzymic activity.

We are studying a protein fraction from wheat flour that makes up about 25% of the gel fraction that remains undissolved when a flour or a dry dough is exhaustively extracted with dilute acetic acid under mild conditions. The protein in the gel fraction represents about 15% of the total protein of the starting flour. So far little has been learned about this fraction, which appears to be part of the glutenin protein, because it is difficult to handle and is insoluble. We have learned that the fraction becomes partly and progressively soluble when a dough is mixed severely, and that its quantity varies among flours of different baking performance. We have found solvents that separate most of the carbohydrate from the gel fraction protein, hence our earlier supposition that this protein was combined in some way with carbohydrate was not sustained. We are now able to prepare a protein fraction that has only a small amount of the carbohydrate xylose included. By column chromatography and gel electrophoresis on agarose, preliminary separations of individual gel fraction proteins are being made. When enough material has been separated, chemical analysis and measurement of physical properties of the separated components will be made.

A related study of the soluble pentosans associated with proteins of wheat flour is being conducted at the Swiss Federal Institute of Technology at Zurich under a P.L. 480 grant. The chemical linkages between pentosans, proteins, and polyphenolic constituents are being characterized. Two approaches are being used. In one, a highly active and specific xylan-splitting enzyme that has been obtained and purified will be used to degrade the pentosan chain that is assumed to be the backbone of the wheat glycoprotein of interest. In the second approach, model compounds are being

prepared that are expected to be similar to the natural glycoprotein. When starch and caffeic acid were used, a high degree of esterification was achieved (i.e., every third glucose unit of the starch was esterified with caffeic acid). The high degree of esterification resulted in a loss of water solubility, so the capability of this model compound to form a gel could not be tested. Similar compounds with lower degrees of esterification will be prepared, and also model compounds in which polyphenols will be esterified with galactomanan and arabino xylan (a natural constituent of flour). Ferulic acid, also a natural constituent of flour, will be used in place of caffeic acid in future research.

The Research Association of British Flour Millers at St. Albans, England, is supported by a P.L. 480 grant to study means of solubilizing gluten proteins so their component parts may be better understood. They have shown that a cupric-bisulfite reagent can be used to solubilize gluten, and they are now studying the effects of this treatment on the proteins. Although they managed to separate the sulfitolized flour proteins, the separated components had a strong tendency to reaggregate. Considerable effort has been spent on finding reagents that will reduce the tendency, which interferes with characterization of separated components. Measurements have been made of the disulfide and thiol groups that are accessible for reaction with sulfite on the separated components. There was a greater accessibility of disulfide groups in a strong-bread making flour than in a weak flour.

P.L. 480 research funds are supporting another attempt to study insoluble gluten by application of ultrasonic vibrations to suspensions of the protein. The work is being conducted at the National Institute of Agronomic Research in Paris, France. An interesting side issue has developed out of this research: they have found that ultrasonic treatment of flour suspensions results in dough formation. One wonders whether the properties of dough formed by ultrasonic vibrations would differ from those in which mechanical mixing is used, and whether some commercial application of ultrasonic treatment would be valuable in continuous dough mixing. In this research, the somewhat different procedure for separating soluble from insoluble proteins produces results that agree with results obtained by other methods, and thus the foundation for further development of this unique method of separation is sound.

A basic investigation of the nitrogenous components of wheat germ is being conducted at the University of Bologna, Italy, supported by P.L. 480 funds. A thorough study of the compositional features of proteins and lipids in wheat germ is underway. Prolamines and glutelins have been found in commercial wheat germ; their presence could be the result of a failure to separate completely the germ tissue from the endosperm during milling. However, in gel electrophoresis of the germ protein preparation, a single albumin component has been reported. Since the endosperm has a number of albumin components, verification of this albumin separation may indicate a suitable method for isolation of germ from endosperm and also may reveal a notable contrast between the proteins from the two portions of the wheat kernel.

2. Maturation of Wheat Flour and Related Chemical Research. Some types of flour, particularly those made from hard red winter wheat, require the addition of oxidative improvers or lengthy storage periods for natural maturation. Oxidative improvers have been found that are widely regarded as safe. Even so, regulations in some continental European countries prohibit the treatment of food materials with almost any chemical agent, so the flour-maturing oxidizers common in America and the United Kingdom are not allowed. Such regulations hinder the development of commercial markets for our most abundant wheats. We are engaged in research aimed at achieving a better understanding of the maturation process, with the long-range objective of finding acceptable alternative methods for maturing wheat flour.

In contract research at Kansas State University, a number of varieties of hard red winter and hard red spring wheats are being studied for chemical composition and responsiveness to potassium bromate and other maturing agents to improve baking strength. Eight hard red winter and five hard red spring varieties from both 1963 and 1964 crops have been tested. The 1963 hard red winter wheats required more bromate, ascorbic acid, and azodicarbonamide than did the spring varieties. However, with potassium persulfate and acetone peroxides, responses were essentially equal. Such results suggest that the improving effects may involve more than one mechanism. Data indicate that hard red spring wheats contain more total lipids than do the hard red winter wheats, with the principal difference being in nonpolar lipids.

We have demonstrated that amino acids form tightly bound complexes with lipid components and metallic ions. This finding supports the hypothesis that metallic ions participate in lipid binding in bread doughs. We have observed that volume changes accompany the formation of such complexes. The volume change should be a useful measurement for estimating the amount of these metallic ions in solution and the stability of the complex.

Oxidative enzymes appear to be related to the mechanism of flour aging. We are supporting contract research at the University of Wisconsin to detect the presence of various oxidation-reduction and other enzyme systems. A wide variety of oxidizing and reducing enzymes have already been shown to be present in whole wheat, in flour, and in several mill fractions. Lipoxidase is the one enzyme that has shown the greatest amount of activity. This enzyme is responsible for oxidizing lipids which are of importance in their chemical relationships with proteins and their effects on maturation of flours and mixing quality of doughs. When the activity of lipoxidase is high, we find that mixing tolerance is low, with the result that the dough breaks down quickly. If these preliminary findings are found to be general, lipoxidase measurement could become an important procedure for describing flours in the specifications from which they are purchased. Quantitative analysis of nine dehydrogenase systems have been run on whole wheat and mill fractions of two hard red spring and two hard red winter wheat varieties. Similar assays were carried out on oxidative enzymes of these samples, including peroxidase, catalase, polyphenol oxidase, and cytochrome oxidase.

Peroxidase and catalase activities were particularly high in all wheats. The spring wheats, however, contained nearly twice as much oxidative activity as did the winter wheats. Data on the dehydrogenases showed a similar trend, so that the two spring wheats tested had a higher overall enzyme activity than did the two winter wheats.

The lipid compositions of whole wheat, ground wheat, flour, bran, and other milling fractions are under investigation at the French School of Milling in Paris, supported by a P.L. 480 grant. The fatty acid composition of wheat lipids in a number of market classes of United States wheats is being analyzed. Further, the effects of wheat lipid components on dough-mixing characteristics are under investigation. To date much of the work done under this grant has been concerned with improving analytical methods and obtaining compositional data on the free fatty acids of the various wheats and their mill fractions. Preliminary correlations of Alveograph resistance measurements with composition of flour indicate that if the total triglyceride lipids are removed from the flour dough, resistance increases considerably. If the lipids are added back to the flour, resistance properties are nearly recovered. The free fatty acids and the acetone-soluble part of the lipids do not affect the resistance, but the unsaponifiable lipids and the acetone-insoluble matter increase it. If just the acetone-soluble and the insoluble parts are added back to defatted flour, higher Alveograph resistance is observed than if all the extracted lipid material is added back. If the dough is allowed a 20-minute rest period between mixing and testing with the Alveograph, results are not all the same as those observed without resting.

Under a P.L. 480 grant the National Institute for Agronomic Research in Paris, France, is conducting a study of the lipase activity of wheat in relation to water-vapor tension, partial pressure of oxygen, and temperature. Lipase activity by some measurements increases appreciably during germination. The advance of knowledge on lipase activity in wheat and on the evolution of lipase action in germinating wheat is hampered by the low intensity of lipasic activity and by a lack of specificity in the analytical methods that have been developed. Therefore, work on this project has been largely centered on studying chemical methods and adapting them so as to obtain greater sensitivity and, therefore, more valuable data. New microanalytical methods using thin-layer chromatography and gas chromatography are being developed. Free fatty acids released from wheat substrates by lipase activity were found to be in the same proportions as those existing in olive oil glycerides. The maximum lipase activity in germinating wheat is located in coleoptile tissues. Calcium ions are essential to the lipase activity. Proteins were extracted from wheat seedling coleoptiles but only a relatively low yield (10-20%) of the lipase activity was preserved in the protein isolation. Perhaps protein-splitting enzymes were active during the extraction procedure; they could have split the lipase protein and rendered it inactive.

The relationship of wheat flavans to various enzyme systems present in wheat flour is being studied under a P.L. 480 grant at the Agricultural Higher School in Poznan, Poland. Accurate methods for extracting and measuring the

flavans of wheat have been developed and an instrument for flavan determination has been designed and constructed. Equipment available previously for fluorometric analysis was not applicable to tightly bound flavan. The new instrument is capable of precise determination of minute flavan fluorescence in an abundance of interfering substances. The total flavan contents and the free riboflavin ratios have been estimated for two spring and four winter wheats and the flour from them. With the new or improved analytical techniques, studies will now be conducted on the influence of flavans on some enzymes of technological importance in flours, and on the effects that storage of wheat and flour has on flavan components.

Another project supported by P.L. 480 grant funds at the National Institute of Agronomic Research in France concerns the behavior of wheat proteins when they are associated with phosphorus-containing compounds. It has become apparent recently that phosphorus-containing fats (phospholipids) can interact with proteins to form new compounds having properties greatly different than those of the uncombined proteins. Phosphorus-containing phytic acid often forms complexes with proteins. With animal proteins, such complexes are almost completely insoluble, but phytic acid complexes with wheat proteins are acid-soluble. This finding may shed light on the properties of wheat gluten, which is insoluble. Studies of the reactions of gluten with phospholipids and phytic acid may be helpful in gaining a better understanding of how gluten acts in forming doughs, batters, and pasta.

B. Flavor

1. Chemistry of Bread Flavor. We are investigating the chemistry associated with bread flavor to find means for enhancing and stabilizing flavor so that bread and other baked products will be more satisfying and desirable for consumers. By smelling the effluent from a gas chromatographic column during analysis of a stable bread-aroma concentrate, we have detected three fractions (only one of which was in sufficient quantity to create a peak on the chromatogram) that have cracker-like aromas similar to that reported as resulting from the reaction of proline and dihydroxyacetone. When we reacted proline with glycerol a similar aroma was produced. Both of these reaction mixtures and the naturally occurring aroma components all separate into three fractions having identical gas chromatographic retention times. A tentative chemical identification has been designated for all three fractions. Baking experiments were conducted with doughs to which the reaction mixture of proline and glycerol was added. The addition improved flavor and aroma of the bread.

Studies of volatile constituents in cell-free prefermented broths revealed eight different alpha-keto acids that almost certainly contribute to the overall aroma produced in commercial bread baking. Three of the acids in the preferment increased with time of incubation. The other five occurred in very small amounts so that quantitative determinations were not reliable.

2. Instability of Wheat Product Flavors. Research on food supply for fallout shelters is funded by transfer from the Office of Civil Defense,

Department of Defense. This research includes a basic investigation of chemical changes involved in oxidative breakdown of fats in cereal products, intended to provide a foundation for development of measurements of food deterioration that will be useful in surveillance of food supplies. New insight gained in the mechanism of autoxidation of methyl linoleate is providing guidance to what actually occurs during oxidation of fats in foods. Results being obtained now at low oxidation levels and at ordinary temperatures are more appropriate models of changes that occur in foods than were the accelerated aging tests used in the past. The primary products of lipid autoxidation, such as aldehydes, are subject to further reactions, for example, oxidation to acids and condensation to trioxolane. Therefore, the primary products are unsatisfactory indicators of the progress of rancidification, and their concentration in the gas space of a food container is not a reliable objective measure of rancidity. Unreactive compounds, such as saturated hydrocarbons, carbon dioxide, and carbon monoxide, which are also products of autoxidation, may furnish even better objective tests, if their development is found to be well correlated with rancidity as determined by organoleptic means.

C. Color, Texture and Other Quality Factors

1. Color. Contract research at Oregon State University is conducted to determine the nature of the color substances in wheat bran and aleurone tissues. A series of phenolic compounds in free and combined state have been found that appear to be the coloring material of the bran. Spectroscopic investigation revealed simple phenolic patterns consisting of mono- and di-substituted aldehydes and acids. The polymeric materials belong to a chemical class known as phlobaphenes. Extracts of red brans from three genetically pure wheat varieties, which were different in intensity of red color, showed differences in both the amount and the nature of isolated materials. These basic studies are expected to provide a foundation for developing decolorizing procedures to use in making whole kernel products and products derived from wheat bran and other milling fractions.

We are also conducting in-house research on bran pigments in connection with studies of proteins that can be extracted from bran. In bran there are two distinct types of pigments, one yellow and one brown. The brown is more closely bound to protein. There is also a clear distinction between protein that adsorbs or binds brown pigment and protein that doesn't. We are now attempting to isolate reasonably pure protein of the type that binds brown pigment in order to determine the nature of the binding.

2. Dough Characteristics. The properties of bread doughs change with mixing; we are studying the chemical reactions involved. The sulfhydryl groups (free sulfur hydrogen terminal groups) of flour proteins appear to be more important to the physical properties and baking behavior of doughs than their low concentration would lead us to expect. Sulfhydryl contents decrease when flours or doughs are matured by oxidation, but sulfhydryl groups must be present during dough mixing if the dough is to have normal elasticity and

extensibility. Doughs mixed in air, so that some sulfhydryl groups are oxidized, differ in physical properties from doughs mixed in nitrogen. Earlier it was believed that the sulfhydryl level did not change significantly when doughs were mixed under nitrogen so that oxidation was prevented. However, we have found that the titratable sulfhydryl groups decrease rapidly in the first two or three minutes of mixing and then return to the original level in 5 to 10 minutes. If the free lipids are removed from the flours by solvent extraction, the initial rapid decrease is eliminated. With longer mixing, sulfhydryl content increases slowly in all flours. Mechanical action in dough mixing is required for such increases, which have been observed to be as much as 50% above the initial flour level. Formation or unmasking of sulfhydryl groups may be the explanation of the reversals in the effects of chemical oxidizing agents and sulfhydryl blocking reagents on the freeing of sulfhydryl groups by dough mixing. Hydrogen sulfide has been detected in the nitrogen atmosphere above doughs when they are protected from air during mixing. The amount of hydrogen sulfide evolved depends upon the mechanical energy applied to the doughs. No hydrogen sulfide has been released by stirring flour water suspensions. If sulfhydryl blocking reagents (iodate or bromate or yeast) are added to the doughs, the amount of hydrogen sulfide released is smaller.

In contract research conducted at Washington State University, the fate of extractable proteins is being followed by use of radioactive tracers. By growing wheat in the presence of radioactive carbon dioxide, a certain amount of radioactivity is incorporated into the wheat kernel and the flour that is obtained from it. Examination of the wheat protein by starch gel electrophoresis indicates that dough mixing transfers radioactive tracers from the slow-moving proteins to the fast-moving proteins, which indicates that protein has been degraded in the mixing process. Possibly fast-moving proteins (albumin or globulin) are released from a state of being bound to slow-moving proteins (gliadins or glutenin). The binding could very well be by disulfide bonds which can be ruptured by mixing.

Basic studies on the solubility of wheat gluten proteins in aqueous systems and on correlations between protein components and baking quality of flours are being conducted under a P.L. 480 grant at the National Center for Scientific Research at Montpellier, France. Three varieties of wheat flour that differ in baking quality have been tested. Although the maximum solubility in water of the glutens was approximately the same for all three flours, the rate of solubilization seemed to correlate with the baking quality--the greater the rate of solubilization the better the baking quality.

At the British Baking Industries Research Association in Chorleywood, England, the effects upon baking quality of variation in wheat flour lipid are being investigated under a P.L. 480 grant. The extractability of lipids from flours from two wheat varieties having different protein contents and different baking quality was studied. Differential extraction of various lipid components with solvent is affected by the amount of water present. Certain binding sites of lipid-to-protein appear to have higher affinity than others.

In a strong wheat flour, 67% of the lipid was associated with gluten proteins, 22% with starch, and 11% with water solubles. Chemical treatment of flour with an agent which causes loss of cohesion in gluten did not change the yields of free and bound lipid.

In the absence of salt, a fat added in dough making increases the binding affinity of phospholipids. With optimum oxidation of dough, addition of fat improved the baking quality still more. The added fat had no effect on the gas retained by a dough at 80° F. or lost at 130° F., and the fat exerted its improving effect in the absence of yeast when glucono-delta-lactone was used as a raising agent. In a study of baking processes that occur in the oven, no evidence was found that fat affects denaturation of protein. In addition to improving loaf rise, fat present in the dough was responsible for a more rapid loss of moisture in the crust, compared with loaves made without added fat.

Interactions of nonfat dry milk and α_s -casein on dough and bread qualities are being investigated by employing a brew fermentation and a Do-Corder to simulate continuous mixing. With brews containing no flour or only 10% flour, 6% nonfat dry milk consistently caused decreases in crumb score and loaf volume, as expected. When 0.28% α_s -casein replaced 4% of the dry milk, adverse effects were no longer observed. A volume increase usually was obtained when the α_s -casein was hydrated separately from the brew and added at the dough-mixing stage. In contrast, with brews containing 30% flour, α_s -casein in the brew had no effect unless hydrated separately, in which case it gave a small volume decrease. The decrease, however, was not as great as that caused by an equivalent amount of nonfat dry milk. From these results it appears that the action of a protein-splitting enzyme in the flour on α_s -casein from the nonfat dry milk may be of only minor importance to continuous-mix bread production. The commercial procedures in continuous-mix bread making are trending towards a higher percentage of flours in brews (as much as 70%). The relatively poor behavior of either isolated α_s -casein or nonfat dry milk in brews containing only 30% of the formula flour indicates that a problem exists that should be studied further.

3. Dough Rheology. The rheological properties of wheat flour doughs play a major role in determining the properties of baked products, so numerous attempts have been made to find rheological testing procedures that correlate with baking quality. Such instruments as the Farinograph and Extensograph produce characteristic curves that correlate with various aspects of flour quality and thus help to improve control over the operation of mills or bakeries. But these instruments are empirical in principle and they measure a complex interaction of several factors, so changes in baking technology (including high-speed mixing, mechanical dough development, continuous processes, etc.) as well as the varying reactions of chemical improvers complicate the problem. Furthermore, sample geometry and loading patterns are not well defined, and these factors influence the experimental results to the extent that they cannot be meaningfully compared with other measurements.

The aim of a fundamental study of dough rheology is to describe macroscopic rheological phenomena in terms of the underlying molecular structure in a bread dough. To do this, we try to observe fundamental stress-strain relationships and compare them with the stress-strain predicted from an assumed molecular architecture. Stress-strain relationships can be measured in terms of elongation or resistance to shear forces. It is difficult to maintain constant sample geometry for measurements of elongation because of gravitational forces on the sample, and it is impossible to achieve pure elongation without introducing some form of shear. Non-uniformities of stress throughout the sample are a further disadvantage. Shear measurements have the advantage of maintaining constant sample geometry so that, for all equivalent points in the material, stress and strain are homogeneous. In dough rheology, a static approach has been used almost exclusively, but a complete description of rheological behavior must be made over a wide time scale. To cover this, dynamic measurements are preferable but because of instrument complexity, they have generally been avoided. The availability of electronic apparatus and electromechanical transducers to refine measurements and computers to solve mathematical equations of physical systems have been responsible for the recent advances in rheological theories. By necessity, the mathematics are very complex. It is only with a very sophisticated level of rheological inquiry that the complexities of dough rheology are likely to be delineated.

Basic investigations on dough rheology are being conducted by contract at Stanford Research Institute in Menlo Park, California, and by P.L. 480 fund grants to the Rheological Laboratory of the Israel Institute of Technology in Haifa and to the Bread Research Institute of Australia in North Ryde, New South Wales.

In Australia a suitable research instrument has been designed and constructed. A dough sample is sheared by application of a sinusoidal varying force that causes the inner of two concentric cylinders to oscillate. Dough samples in the annular space between the cylinders resist the oscillation, and measurement of this resistance provides data on the viscoelastic properties of the samples. When very small strains are induced, the physical properties of the dough are not influenced by deforming stresses, that is, the behavior is linear. At larger strains, however, the effects are not linear. In previous work of this type, only larger strains were imposed, hence development of a mathematical model was seriously hampered by the nonlinearity of the stress-strain formulas. The complicated effects of temperature, water absorption, and oscillation frequency on viscoelastic functions is being investigated. Preliminary investigation of oxidative improvers indicate that they affect the elastic property much more than the viscous property. This interesting lead will be pursued.

In closely related studies at Stanford Research Institute, large deformation and failure properties are being investigated by pulling dough rings on an Instron tester. Rings with a horizontal pull are less satisfactory than those with a vertical pull, because of the nonuniform friction between horizontal

dough rings and the tray that supports them. The vertical method too, has errors which are introduced by the lack of free slippage over the hooks holding the rings and the sag of the rings under their own weight, but these problems can be largely corrected for. Stress-strain data obtained early in this work showed considerable scatter, but improvements are contemplated in which the dough rings will be immersed in a fluorocarbon or silicone oil of density equivalent to that of dough so that buoyant effects will eliminate sagging of specimens. Dynamic methods for testing linear viscoelastic behavior are also being studied, and equipment is being designed and constructed similar to that available for other types of polymeric materials. Dough differs from many viscoelastic materials in that it can be tested over only a very limited temperature range; other substances can be tested over a wide temperature range at limited frequencies of dynamic stress. To obtain equivalent information for doughs, the measuring instrument must have a much wider frequency range.

In the project in Israel, very rapid measurements are made of tensile stress and strain relationships, relaxation times, and elastic recovery behavior in dough specimens taken at intervals during the dough-mixing cycle. Instruments for rheological measurement were redesigned, then data were obtained to evaluate possible correlations with Farinograph and Brabender Extensograph measurements. Measurements that separated the elastic and viscous components of dough deformation led to a revision of the tentative mathematical model of wheat flour dough. Data will now be obtained on flours from four varieties of wheat supplied from the United States.

The group in Israel also designed new instruments for measurement of viscoelastic properties of dough. One of them, a constant-velocity extensometer, measures dough at high stresses and rates of strain for comparison with the Brabender Extensograph. With this equipment, viscous and elastic deformations can be determined over a much wider range of loading times than usual; these data can be related to the decay of elasticity in dough. In order to widen the range of viscoelastic properties of dough, additives known to affect mechanical properties of dough have been used. Although knowledge of dough properties has been materially advanced in this work, the correlations between these properties and the baking quality of flours are still somewhat obscure. A good foundation is being laid, however, for an ultimate solution of the rheological problems of dough mixing and baking.

D. Microbiology and Toxicology

1. Microbiology of Wheat Food Products. The heat of baking destroys most of the contaminating microorganisms in conventional bakery foods, but when a flour is to be used in foods such as meat pies, soups, and biscuits intended for frozen or refrigerated storage, a low microbial population is highly desirable. If such products are improperly handled, spoilage may result. Contract research on means for reducing microbial contamination of milled flour has been concluded at the American Institute of Baking in Chicago. Four different types of microorganisms (Bacillus subtilis, Aspergillus flavus,

Staphylococcus aureus, and Escherichia coli) were used, individually, to inoculate flour at the rate of 1 million per gram. Frozen pie dough and soup prepared from these flours were tested at -3° F. and at 20° F. The higher temperature represents grossly unsatisfactory storage conditions for frozen products. Biscuit dough was stored at 37° F., representing good storage temperature, and at 37° F. cycled once a day to 70° F. for 6 hours to represent grossly unsatisfactory conditions of storage. At the unsatisfactory storage temperatures, pie dough became rancid; soup underwent a drastic change in consistency and browned excessively; refrigerated biscuits lost volume when baked, the crumb became gummy and gray, and an off-odor developed. Stability of frozen products was very materially reduced by the use of inoculated flour as compared with uninoculated flour. Inoculation did not materially affect stability of the biscuit doughs, but the storage defects were drastic even with flour that had not been inoculated. Propylene oxide can be used to reduce microbial contamination, but a more desirable control of micro-organisms was found to be a 45-minute heat treatment of flour at 130° C. Heating at this temperature is nearly as effective as treatment with propylene oxide at levels of 100 parts per million. The cost of propylene oxide, its flammability, and the possibility of an odorous residue lead to the conclusion that heat treatment of flour is better than the chemical treatment.

E. Technology--Process and Product Development

1. Bulgur, WURLD Wheat, and Related Products. The capacity for food production in North America that exceeds local population needs is a weapon for combating hunger in the developing nations of the world. The surplus conditions are expected to continue for two decades or more, which may provide sufficient time for agricultural advances that hopefully will make the developing nations more self-sufficient. The North American surplus productive capacity is for wheat, but most of the food-deficient areas of the world do not have a tradition of eating wheat or wheat products.

A new product developed at the Western Lab, WURLD wheat, may be a form that is acceptable in the areas of greatest need. It is a whole kernel or cracked kernel product from which the bran has been removed by lye-peeling. A 2500-lb. lot of WURLD wheat shipped to Hong Kong over a year ago for pilot acceptance tests by voluntary welfare agencies was sufficiently successful to warrant further acceptability and marketability tests in Hong Kong. Pilot plant operations were scaled up to produce at a level of about 200 lbs. per hour. Now a 3-ton lot of WURLD wheat has been sent to Hong Kong for testing under the auspices of the Church World Service. We hope to get data on commercial marketability as well as consumer acceptance from this second trial. Two companies have conferred with us about the WURLD wheat peeling operation; one company can produce WURLD wheat at a rate of 2 tons per hour, the other at 25 tons per hour. Stability studies of WURLD wheat indicate that it has sufficient shelf life to enter distribution channels like those used for bulgur.

Both gun-puffed bulgur and WURLD wheat have a pleasant mildly toasted taste and they are easily chewed. WURLD wheat puffed in hot air is more tender and

friable than the product obtained from bulgur, and it is much better for use in confectionery and bakery products as well as in convenience products requiring rapid uptake of water.

WORLD wheat can carry a high level of high-quality protein when it is coated with soy flour or high-protein fractions obtained from soy beans. Coatings are steamed in place on whole or cracked kernels and then dried. The coating becomes very firmly attached; it does not slough off even when cooked in excess water. We have also added protein to bulgur, rolled wheat, rolled bulgur and rolled WORLD wheat in this way. A protein-enriched rolled wheat or rolled bulgur can be instantly reconstituted into a highly nutritious gruel by mixing it with boiling water. As such it can be a health giving food for undernourished children in regions where protein deficiency abounds.

Stabilities of bulgur wafers and of a line of food adjuncts to make bulgur wafers more palatable are being studied under a purchase contract at Oregon State University in Corvallis. This research is supported by transfer of funds from the Office of Civil Defense, Department of Defense. After the wafers had been stored 40 months, taste panels preferred those made from bulgur that had received an atmospheric rather than a pressure cook and had been compounded with malt syrup rather than corn syrup, and packed in an atmosphere of nitrogen instead of air and stored at 70° F. or lower. Analyses of headspace gas show that more carbon monoxide and carbon dioxide are produced by wafers packed without nitrogen replacement of the air and stored at higher temperatures, and the amount of oxygen absorbed or chemically reacted is greater.

Twelve adjuncts for use with bulgur wafer in rations stocked in fallout shelters have been in storage for 2-1/2 years. Most of them benefit from being packed in a nitrogen atmosphere; only beef soup mix and dry strawberry spread mix seem to survive better in air. Apple topping and wild cherry icing mixes show no differences. In-package desiccant was helpful in maintaining quality in chicken soup, chocolate pudding, and curry sauce mixes. As expected, the lower storage temperatures were generally advantageous. Carbon dioxide in headspace gas paralleled deteriorative change in that more carbon dioxide was found in samples stored at higher temperatures. However, the liberation of carbon dioxide cannot be measured in samples containing an in-package desiccant because it is absorbed by the desiccant. Carbon monoxide measurement, however, may be useful as compositional data that parallel deteriorative processes. Under carefully controlled and reproducible conditions, the measurement of carbon dioxide and carbon monoxide may be useful in determining residual storage life of canned dry products.

2. New Wheat-Based Foods. Using inexpensive dry milling methods, we have concentrated protein of high biological value from millfeeds. In this way, sizable proportions of these low-cost materials can be recovered for food as well as for high-quality feed ingredients. Application of these processes could result in greater financial returns to millers, offer relief of pressure against high prices for flour, and lead to recovery of valuable protein

for use in developing countries overseas. By dry milling and sifting, protein concentrates with low-fiber content have been prepared from wheat mill-feeds, coarse bran, fine bran and shorts--byproducts now used as animal feed. Starch and total sugars, as well as protein, were concentrated in the fine flour-like fraction that passed a 7XX bolting cloth; fiber, ash, and pentosan contents were reduced; fat content remained relatively unchanged. Adjustment of moisture content of the millfeeds before milling greatly affected the yield of flour-like product as well as its protein concentration. Highest yields were obtained at lowest moisture levels, but for most desirable composition and ease in processing conditions, the moisture range of 9 to 11% appears most promising. At this moisture content, 20 to 30% of bran fractions can be recovered as protein concentrate and 50% of shorts. These concentrates have 2 to 4% fiber and, therefore, can be used as human food. Amino acid analysis and protein efficiency ratio evaluations with laboratory animals show that the concentrate from shorts is of good nutritional quality.

Protein concentrates from shorts have been formulated with farina, corn meal, nonfat dry milk solids and soy flour for use as simple gruel products that are palatable and highly nutritious. A new process was derived to prepare protein concentrates from flour by incorporation of soluble soy protein and vegetable oils which modified the gluten recovery from a water slurry. The method has an advantage over conventional wet milling in that a starch by-product can be easily separated from the dispersed gluten and recovered. Simplification of the separation of starch and protein from flour should reduce the cost of this process.

High-protein wheat flours can be converted to milk-like beverages that have bland flavor and light color. They are easily dispersed in water. To improve dispersibility flour or the protein concentrates are digested with starch and protein-splitting enzymes under controlled conditions. The products can be used as beverages and gruels and should be useful particularly in enriching diets of infants and small children in regions of protein deficiency.

3. Carrying Capacity of Hard Red Winter Wheat. To provide information that may lead to wider markets for surplus United States hard red winter wheats, we are investigating the capacity of these wheats to carry the soft low-protein European wheats in the making of high-quality yeast-raised bread. Research on blending characteristics is conducted, by contract at Kansas State University, with samples of soft wheats procured from Germany, England, Sweden, The Netherlands, Austria, France, and Belgium and samples of hard red winter and spring wheats from the United States and Canada. European wheats are being tested for milling, blending, and baking characteristics. In addition, two South African soft wheats have been used in the tests, because their resemblance to European soft wheats provides a better exploratory experiment than would have been possible with United States soft wheats. Even relatively small additions of higher protein hard wheats from the United States improved baking characteristics of soft wheats in regard to absorption, mixing tolerance, loaf volume, and crumb texture. U.S. winter wheats at two

protein levels (10.0 and 11.3%) produced some improvement, but North American spring wheat of still higher protein level gave better results.

Flours from Swedish and Belgian wheats required no malt in the bread formulas, indicating the presence of sprouted or immature wheat--a major problem to European flour mills. High humidity in grain at time of harvest frequently allows some of the wheat to sprout during harvest and storage.

4. Malting Technology. We have discovered that agitating a bed of wheat or barley during the steeping process inhibits sprouting but not enzyme formation. Malting capacity for a given volume of equipment can be increased 50% by agitation, and loss of dry material from the malted wheat is less because no rootlets develop. Such factors as lack of oxygen in the steeping solution, effect of elevated temperatures, and the production of a water barrier beneath the kernel epidermis were found not to be responsible for the sprout inhibition. In normal steeping, a growth-inhibiting substance is eluted from the hulls of wheat and barley, and more of it is produced under agitation. We suggest that diffusion of this substance into the kernel is greater as a result of the agitation. Another hypothesis for the difference between still and agitative steeping is that mechanical damage to the germ may result from agitation.

Contract research is being conducted at the University of Minnesota in St. Paul to study the changes in proteins of wheat during malting. Progress has been made in developing methods for extracting and fractionating nitrogenous constituents of ungerminated and germinated wheat, and a preliminary confirmation of the expected increase of protein efficiency ratio as a result of germination has been obtained. The prospects are encouraging for development of new and improved wheat food products with enhanced nutritive properties.

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WHEAT UTILIZATION - FEED
Northern Utilization Research and Development Division, ARS

Problem. In the last 2 years the use of wheat for feed increased to nearly 100 million bushels per year, more than twice the amount used in any other recent year. Unfortunately, wheat has certain performance drawbacks as a feed. Research that develops new processes to improve feeding quality of wheat will benefit both growers and feeders in wheat-producing areas, since it will place this grain in a more competitive position as compared with other grains, and it can reduce freight costs.

Millfeeds are not used extensively in modern poultry and swine rations because the high fiber content cannot be tolerated in high-energy rations. If inexpensive ways of separating low-fiber, high-protein fractions from millfeeds are developed, these new materials can be used as protein and energy sources for nonruminant diets, and the overall value of milling byproducts will be increased. Flour production is expected to increase in the near future to reflect the demands of our increasing domestic population and of the new export markets which are developing. More milling will result, of course, in more millfeeds. If these millfeeds cannot be utilized efficiently and effectively, the price for flour will have to increase to carry the economic burden.

Meat production, particularly poultry, is increasing rapidly in Japan and the European Economic Community where modern efficient methods have been introduced. This development depresses the opportunity for exporting poultry and other meats into these important trade areas, but it offers an increasing opportunity to sell feeds. Upgrading of wheat millfeeds through utilization research will increase our export markets.

USDA AND COOPERATIVE PROGRAMS

The Department conducts a continuing, long-range program of research involving analytical, organic, and physical chemists, biochemists, microbiologists, systematic biologists, and chemical engineers engaged in basic, applied, and developmental studies pertinent to utilization of wheat in feed.

The Federal scientific effort (Northern region) for research on utilization of wheat in feeds totals 3.8 scientist man-years, of which .9 is devoted to chemical composition and physical properties; 2.1 to microbiology and toxicology; and .8 to technology - process and product development.

Research at Peoria, Illinois, on chemical composition and physical properties (.9 scientist man-year) involves basic investigations of the microscopic and ultrastructure of wheat grains and the effects of various treatments. During the year, research on the effects of wheat conditioning and characteristics of milled fractions was completed.

Research at Peoria, Illinois, on microbiology and toxicology (1.3 scientist man-years) is concerned with studies on the production of mycotoxins by Aspergillus flavus and related molds. Research contracts (.8 scientist man-year*) are in effect with A. D. Little, Inc., Cambridge, Massachusetts, for studies on stabilization of fermentative β -carotene; with Consolidated Laboratories, Inc., Chicago Heights, Illinois, for research on the use of antimetabolites to facilitate selection of higher yielding strains of microorganisms producing β -carotene; and with the Agricultural Experiment Station, South Dakota State University, Brookings, South Dakota, for survey of the genus Aspergillus to find and identify species producing toxic metabolites.

Research conducted at Peoria, Illinois, on technology - process and product development (.8 scientist man-year) is concerned with studies on new techniques of milling and fractionation to obtain improved products and with development of methods for minimizing radioactive contamination of wheat and milled fractions.

The Department also sponsors research in this area conducted under grants of PL 480 funds. Research on microbiology and toxicology involves a grant to the Agricultural University, Poznan, Poland, for studies to increase the yield of β -carotene produced by fermentation of cereal grains. Effort on this project is prorated among corn, wheat, and sorghum. During the reporting period, research was completed at the "Giuliana Ronzoni" Scientific Institute of Chemistry and Biochemistry, Milan, Italy, on production of Vitamin B₁₃ and at the National Institute for Agronomic Research, Paris, France, on mutation of yeasts for improved feeds.

PROGRAM OF STATE EXPERIMENT STATIONS

A total of 3.0 scientist man-years is devoted to this area of research.

PROGRESS -- USDA AND COOPERATIVE PROGRAMS

A. Chemical Composition and Physical Properties

Studies on the microscopic and ultrastructure of wheat are relevant to utilization of wheat in feeds. Results are reported under Area 5, subheading A-1.

B. Microbiology and Fermentation

1. Microbial carotenoids. Research on fermentative conversion of cereal grains to carotenoid-rich additives for feeds is applicable to wheat. Results are reported under Area 3, subheading B-1.

*Work covers more than one commodity; only effort allocated to wheat is included in total.

2. Aflatoxin investigations. Studies on toxins produced by molds are important to utilization of wheat in feeds. Results are reported under Area 3, subheading B-2.

3. Vitamin B₁₃. Studies on Vitamin B₁₃ are pertinent to utilization of wheat in feeds. Results are reported under Area 3, subheading B-3 (PL 480 research).

4. Improved feeds by mutation of yeasts. Studies on mutant yeasts capable of producing high yields of sulfur-containing amino acids are relevant to feed utilization of wheat. Results are reported under Area 3, subheading B-4 (PL 480 research).

C. Technology - Process and Product Development

Research on milling and fractionation of wheat and on reduction of radioactive contamination in wheat and milled products is relevant to utilization of wheat in feeds. Results are reported under Area 5, subheading C.

PUBLICATIONS -- USDA AND COOPERATIVE PROGRAMS

None.

RELATED PUBLICATIONS OF STATE EXPERIMENT STATIONS

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WHEAT UTILIZATION - FEED

Western Utilization Research and Development Division, ARS

Problem. Wheat can now compete with feed grains as animal feed because new legislation on wheat pricing makes it economical. In the last two years the use of wheat for feed increased to nearly 100 million bushels per year, more than twice the amount used in any other recent year. Unfortunately, wheat has certain performance drawbacks as a feed. Research to improve the feeding quality of wheat would greatly benefit both growers and feeders in wheat-producing areas, since it would place this grain in a more competitive position as compared with other grains.

Millfeeds are not used extensively in modern poultry and swine rations because the high fiber content cannot be tolerated in high-energy rations. If inexpensive ways of separating low-fiber, high-protein fractions from millfeeds are developed, these new materials can be used as protein and energy sources for non-ruminant diets, and the overall value of milling byproducts will be increased. Flour production is expected to increase in the near future to reflect the demands of our increasing domestic population and of the new export markets which are developing. More milling will result, of course, in more millfeeds. If these millfeeds cannot be utilized efficiently and effectively, the price for flour will have to increase to carry the economic burden.

Meat production, particularly poultry, is increasing rapidly in Japan and the European Economic Community where modern efficient methods have been introduced. This development depresses the opportunity for exporting poultry and other meats into these important trade areas, but it offers an increasing opportunity to sell feeds. Upgrading of wheat millfeeds through utilization research will increase our export markets.

USDA AND COOPERATIVE PROGRAM

Research at Albany, California, on utilization of wheat seeks to develop new processes to convert milling byproducts into high value feeds and to modify whole wheat so that it is more economical for use as a feed grain. Research on barley is also conducted.

The Federal program of research in this area totals 2.9 scientist man-years, of which 1.4 are assigned to investigations on chemical composition and physical properties and 1.5 are assigned to technology--process and product development.

PROGRAM OF STATE EXPERIMENT STATIONS

A total of 3 scientist man-years is devoted to this area of research.

PROGRESS -- USDA AND COOPERATIVE PROGRAMS

A. Chemical Composition and Physical Properties

1. Wheat Proteins. We are improving the methods for amino acid analysis of wheat products. The hydrolysis step is the major cause of variability of analytical results. Some amino acids (e.g., threonine, serine, cystine and methionine) are partially destroyed during hydrolysis. Other amino acids are liberated very slowly and require excessively prolonged hydrolysis (e.g., valine and isoleucine). The relative rates of destruction of labile amino acids and liberation of other amino acids are uniform for food or feed products but not for pure isolated proteins. A systematic study was made using an accelerated system with an automatic amino acid analyzer to determine effects of time of hydrolysis of wheat and wheat bran on apparent amino acid composition. Correction factors were calculated to permit precise determination, from a single 24-hour hydrolysate, of all common amino acids of wheat except cystine, methionine and tryptophane. A second hydrolysis procedure is used to determine cystine and methionine.

2. Non-protein Components. Progress has been made on methods of analysis for nondigestible holocellulose and lignin in millfeeds. The large amount of starch present interferes with the analysis. We have found it is better to remove most but not all the starch by enzymes and then determine the amount of starch remaining. The procedure is simple compared with enzymic removal of all of the starch. We are also testing and developing simplified methods for analyzing lipids in samples of wheat and mill fractions. Test material is supplied by the Technical Subcommittee on Millfeeds of the Millers National Federation.

B. Technology--Process and Product Development

1. Improved Feeds from Milling Byproducts. Efficient utilization of millfeeds is a major problem facing millers, growers, and a hungry world. Nearly 28% of whole grain wheat is recovered as non-flour byproducts during milling of white flour. These millfeeds have been sold as feeds, and only recently has an intensive research program been undertaken to upgrade both their dollar and nutritional value. Milling byproducts contain important nutrients at levels of concentration even higher than in the milled flour, but because of general variability of quality and condition, they cannot compete for use in mixed feeds with other sources of protein, carbohydrate and other nutrients. The unrealized feed values in mill byproducts adversely affect milling profits and are reflected in higher flour prices as well as in pressure to reduce wheat prices to growers.

We are preparing protein concentrates with low fiber content from wheat millfeeds, coarse bran, fine bran, and shorts by dry milling and sifting. In addition to protein, much of the starch and total sugars from these feed fractions are also concentrated in the fine flourlike fractions. Fiber, ash and pentosan contents, on the other hand, are reduced. Moisture content

of millfeeds before milling greatly affects yield of protein concentrate as well as the concentration of the protein. Highest yields were obtained with low moisture level but, from the standpoint of desirable composition and ease of processing, remilling the millfeed fractions at a moisture range of 9-11% appears most promising. Yields of 20-30% of low-fiber protein concentrate are possible with bran fractions; up to 50% yields are possible with shorts. Fiber content of these products ranges from 2-4%, so they have utility in mixed feeds for non-ruminants and also for human food. Amino acid analyses and protein efficiency ratios show that the concentrate from shorts is of good nutritional quality.

2. Improved Use of Wheat for Feeds. Experiments are underway to improve the nutritional quality of wheats by simple processing treatments. Preliminary information has been obtained on pressure cooking of wheat at various steam pressures, with and without added acid, and on hot-air puffing of whole wheat. Data obtained from tests in an artificial rumen indicate that nutritive value is improved, both by cooking and by puffing. If these data can be verified with large animals under usual fattening conditions, they will indicate that costs of meat production can be reduced significantly.

We are supporting research with P.L. 480 funds at Cambridge University in England to develop rapid chemical methods that can be applied during processing of wheat and wheat products for assay of the biological value of wheat proteins. Our work on improvement in nutritive value of wheat, both for human and for animal feeding, by controlled moisture-heat treatments stems from the observations of this project. Careful cooking and drying of wheat improves its protein efficiency ratio, although other protein quality measurements are less affected. But careful control in processing cereal foods and feeds is necessary, because overheating can impair nutritive quality. It is apparent that the major effects of cooking are changes in carbohydrate and other nonprotein wheat components. Although a rapid chemical method for assay of biological value of wheat proteins has not been developed in this project, a number of useful leads for further research have been obtained.

PUBLICATIONS - USDA AND COOPERATIVE PROGRAMS

Chemical Composition and Physical Properties

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Technology - Process and Product Development

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Milner, C. K., and Woodforde, J. 1965. The effect of heat in drying on the nutritive value of wheat for animal feed. J. Sci. Food Agr. 16(7), pp. 367-373.1/

RELATED PUBLICATIONS OF STATE EXPERIMENT STATIONS

Chemical Composition, Physical Properties and Structure

Attia, F., and Creek, R. D. 1965. Studies on raw and heated wheat germ for young chicks. Cereal Chem. 42(5), pp. 494-497. Md.

Castanera, Esther G., and Hassid, W. Z. 1965. Properties of uridine diphosphate D-glucuronic acid decarboxylase from wheat germ. Arch. Biochem. Biophys. 110(3), pp. 462-474. Calif.

Elnaghy, M. A., and Nordin, P. 1965. The soluble nucleotides of the leaf rust uredospores and the loose smut chlamydospores. Arch. Biochem. Biophys. 110(3), pp. 593-600. Kans.

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Stebbins, G. L., and Jura, P. 1965. Differential synthesis of nucleic acids associated with cellular differentiation in the leaf sheath epidermis of barley. Sci. 150(3694), pp. 385-386. Calif.

1/ Research supported by P.L. 480 funds.

WHEAT UTILIZATION - INDUSTRIAL PRODUCTS

Northern Utilization Research and Development Division, ARS

Problem. Although the principal use of wheat is as food, a total of about 200 million pounds of wheat starch and flour was consumed by industry in 1963. Loss of this market would detract from the economic value of wheat as a crop. As a food grain, wheat commands a price that is generally unfavorable to its utilization as an industrial raw material. However, in certain areas, notably the Pacific Northwest where corn is not grown, wheat is cheaper than either corn or sorghum. Furthermore, new high-yielding strains of wheat especially suited for this area are being developed. There are many paper mills in this area, and the need for technology to use starch and flour produced there has materialized. Much of the starch now used comes from imported tapioca.

Other possibilities for economic and noncompetitive industrial outlets for wheat are based on use of whole ground wheat and millfeeds, including wheat bran, and on exploitation of the properties of wheat gluten protein, which has unique properties not possessed by other cereal proteins.

Not only paper and paper products, but also coatings, adhesives, thickeners, and plastics offer excellent opportunities for industrial products derived from wheat. A more detailed discussion of industrial outlets for cereal starches and flours is given in Area No. 1, Corn Utilization - Industrial Products.

To achieve the objective, research is needed to learn how wheat flour, starch, and milling fractions can best be modified to provide new and improved properties such as water resistance, dispersibility, paste viscosity, tack, and adhesive bond strength. The possibilities of achieving some of these improvements by modification of the gluten component of flour should be investigated. Conditions must be established for optimum use of industrially promising products now under study such as acid- and enzyme-modified flours and xanthated bran and millfeeds. Basic research should provide leads to other products and processes for future development.

USDA AND COOPERATIVE PROGRAMS

The Department conducts a continuing, long-range program of research involving analytical, organic, and physical chemists, biochemists, microbiologists, systematic biologists, and chemical engineers engaged in basic, applied, and developmental studies on the composition of wheat, on characterization and properties of the components, and on their chemical and microbiological conversion to useful industrial products.

The Federal scientific effort for research on industrial utilization of wheat totals 54.8 scientist man-years. Of this number, 9.2 are devoted to

chemical composition, physical properties and structure; 19.1 to chemical and physical investigations to improve products; 13.8 to microbiology and fermentation; and 12.7 to technology - process and product development.

Research at Peoria, Illinois, on chemical composition, physical properties and structure (7.6 scientist man-years) involves study of wheat flour, starch, and the component proteins of wheat gluten. Research on wheat starch is integrated with that on corn and sorghum starches. The work on wheat includes study of the microscopic and ultrastructure of wheat grains and flours and of changes induced by various treatments. A contract (.6 scientist man-year) is in effect with Purdue Research Foundation for studies on alkaline desulfurization of wheat gluten proteins. Grants (1.0 scientist man-year) have been made to Marquette University, Milwaukee, Wisconsin, for basic studies on intermediates involved in forming glycoprotein linkages; to Iowa State University, Ames, Iowa, for basic research* on heat, mass, and momentum transport of cereal starches and flours; and to Purdue Research Foundation, Lafayette, Indiana, for research* on the effects of disulfide bond cleavage on the structure of corn and wheat endosperm proteins.

Research at Peoria, Illinois, on chemical and physical investigations to improve products (14.0 scientist man-years) includes study of the chemical reactions of wheat starch, flour, protein and milling fractions with the objective of discovering new chemical products and processes having potential for industrial use. Research on wheat starch is integrated with that on corn starch. During the year, one phase of this work involving study of possible means for preparing amino acid derivatives of starch was completed and replaced by research on synthesis of halogen derivatives of starch. Also, a project on hydrophilic derivatives of wheat flour was completed. Acid-modified flour, one of the products developed under this project, is being further evaluated under a research contract. Research contracts (2.8 scientist man-years) are in effect with the Arizona Agricultural Experiment Station, University of Arizona, Tucson, Arizona, for basic studies* on the reaction of acetylene with methyl glucoside; with The Johns Hopkins University, Baltimore, Maryland, for basic research* on the reactions of starch in fluid dynamic media; to the University of Pittsburgh, Pittsburgh, Pennsylvania, for studies on dielectric activation of starch; with Southern Illinois University, Carbondale, Illinois, for investigations* on synthesis of maltooligosaccharides; with Stanford Research Institute, Menlo Park, California, for research* on graft copolymers of cereal starches with vinyl-type monomers; with the Institute of Paper Chemistry, Appleton, Wisconsin, for investigation* of physical chemical factors affecting retention and effectiveness of starch xanthates and xanthides in paper; and with IIT Research Institute, Chicago, Illinois, on preparation, characterization, and chemical modification of polypeptides

*Work covers more than one commodity; only effort allocated to wheat is included in total.

derived from wheat gluten. Contract research was completed by Ohio State University, Columbus, Ohio, on synthesis of amino derivatives of starch and by the University of Arizona Agricultural Experiment Station, Tucson, Arizona, on the reaction of starch with mercaptans. Grants (2.3 scientist man-years*) have been made to Ohio State University Research Foundation, Columbus, Ohio, for basic research on the reaction of vinyl ethers with carbohydrates; to Ohio State University, Columbus, Ohio, for basic investigations of unsaturated and sulfur-containing carbohydrates and of the amination of starch; to Purdue Research Foundation, Lafayette, Indiana, for studies on sugars containing carbon-bound nitrogen, phosphorus and sulfur; and to the University of Arizona, Tucson, Arizona, for basic research on the reaction of starch with diepoxides.

Research on microbiology and fermentation conducted at Peoria, Illinois (11.7 scientist man-years), includes studies on the use of microorganisms to convert cereal-based media to industrially useful products such as chemicals, enzymes, polymers, and biological insecticides. This research is integrated with similar studies based on corn. A large collection of pure cultures of industrially and agriculturally important microorganisms is maintained. The Pioneering Laboratory for Microbiological Chemistry conducts research on microbiological reactions and products. Investigations on biological insecticides for Japanese beetle and on other insect control agents is cooperative with Entomology Research Division and Plant Pest Control Division. Research on plant antibiotics involves cooperation with Crops Research Division. Research contracts (.7 scientist man-year*) are in effect at Michigan State University, East Lansing, Michigan, for basic research on enzyme activity in sporulation; at Kansas State University, Manhattan, Kansas, for investigation of stabilization of vegetative cells of the pathogenic organisms; at the University of Minnesota, St. Paul, Minnesota, for fundamental studies on the transfer of genetic determinants of sporulation from one microorganism to another; at Baylor University, Houston, Texas, for investigation of morphological changes involved in sporulation; and at the American Type Culture Collection, Rockville, Maryland, for studies on preservation of certain microorganisms for which lyophilization is ineffective. Contract research at the University of Illinois, Urbana, Illinois, on the applicability of a sporulation factor produced by bacteria to Japanese beetle pathogens has been completed. Grants (1.4 scientist man-years*) have been made to Cornell University, Ithaca, New York, for fundamental studies on biphasic fermentation; to the Nebraska Agricultural Experiment Station, University of Nebraska, Lincoln, Nebraska, for investigations on the nature of amylase enzymes; to Kansas State University, Manhattan, Kansas, for investigations on separation of enzymes and proteins by disc electrophoresis; to Iowa State University, Ames, Iowa, for investigation on bacterial amylases and their action

*Work covers more than one commodity; only effort allocated to wheat is included in total.

patterns; to the University of Wisconsin, Madison, Wisconsin, for studies on the fine structure of polysaccharide B-1973; and to the University of Arkansas, Fayetteville, Arkansas, for investigation of the mechanism of enzymatic hydrolysis of starch.

Research conducted at Peoria, Illinois, on technology - process and product development (5.8 scientist man-years) is concerned with detailed study and evaluation of wheat-derived products having definite potential for industrial utilization and of processes for making them. Also, studies are conducted on modified techniques for milling and fractionating wheat to obtain improved materials for industrial and other purposes. Research involving chemical modification of wheat starch is integrated with that on corn starch. Research contracts (6.9 scientist man-years) are in effect with Stanford Research Institute, Menlo Park, California, for process development* of selected starch graft copolymers; with Battelle Memorial Institute, Columbus, Ohio, for developmental research* on starch and other cereal grain xanthides, for development of optimal processes for incorporating wheat-derived xanthides into paper products, and for studies* on starch derivatives for use as colloids in water-emulsion paints; with Western Michigan University, Kalamazoo, Michigan, for evaluation* of modified cyanoethylated starches for applications in paper; with Archer Daniels Midland Company, Minneapolis, Minnesota, for investigations* on the use of starch glycosides in coatings and plastics; with the Brown Company, Berlin, New Hampshire, for evaluating acid-modified flour as a paper size; and with the University of Akron, Akron, Ohio, for evaluation* of starch and starch derivatives as reinforcing agents for natural and synthetic rubber. During the year, contract research on evaluation of starch polyol urethane foams was completed by Archer Daniels Midland Company, Minneapolis, Minnesota, and studies on pneumatic fluidization of wheat flour were completed at Iowa State University, Ames, Iowa.

The Department also sponsors research on cereal starches conducted by foreign institutions under grants of PL 480 funds.** Research on chemical composition and physical properties involves grants to the University of London, London, England, for research on debranching enzymes and their use in studying the fine structure of starch components (5 years, 1963-1968); University of Osaka Prefecture, Sakai, Japan, for development of an analytical method for carbonyl groups in carbohydrates (4 years, 1964-1968); and to "Giuliana Ronzoni" Scientific Institute for Chemistry and Biochemistry, Milan, Italy, for research on glucopyranose rings in starches and dextrans (5 years, 1962-1967).

Research on chemical and physical investigations to improve products involves grants to Hebrew University, Jerusalem, Israel, for studies on

* Work covers more than one commodity; only effort allocated to wheat is included in total.

**Effort prorated among corn, wheat, and grain sorghum.

starch vinyl and epoxide graft copolymers (4 years, 1963-1967); National Institute of Technology, Rio de Janeiro, Brazil, for research on phosphorus- and sulfur-containing cationic starches (5 years, 1962-1967); Ahmedabad Textile Industry's Research Association, Ahmedabad, India, for research on starch-gum copolymers prepared by codextrinization (5 years, 1963-1968), and for studies on preparation and characterization of hydroxyethyl ethers of cereal starches (5 years, 1965-1970); Academy of Sciences and Chemical Institute "Boris Kidric," Ljubljana, Yugoslavia, for studies on modification of starch by moisture and temperature treatments (5 years, 1964-1969); Plastics Research Institute TNO, Delft, The Netherlands, for research on preparation of metal alkoxides of starch for use as intermediates in synthesis (5 years, 1964-1969); University of Edinburgh, Edinburgh, Scotland, for studies on the mechanism and structural changes involved in thermal, acid, and alkaline degradation of starches (5 years, 1964-1969); and to the Institute for Fibres and Forest Products, Jerusalem, Israel, for studies on the mechanism and products of mild oxidation of starch (5 years, 1963-1968). During the year, research was completed on fatty chemical derivatives of starch dextrans at the Institute of Industrial Chemistry, Bologna, Italy, and on changes induced in starch by gamma-irradiation at the National Institute of Agronomic Research, Paris, France.

Research on microbiology and fermentation involves grants to the University of Milan, Milan, Italy, for basic studies on the metabolic pathway to 5-ketogluconic acid in Acetobacter species (5 years, 1961-1966); University of Allahabad, Allahabad, India, for collection of new Mucorales species (5 years, 1961-1966), and studies on survival of lyophilized microorganisms (5 years, 1962-1967); University of Newcastle upon Tyne (formerly University of Durham), Newcastle upon Tyne, England, for investigations of sugar phosphate derivatives in molds (5 years, 1962-1967); Central Drug Research Institute, Lucknow, India, for studies on aerobic actinomycetes in India to find new accessions for the ARS Culture Collection (5 years, 1965-1970); to the University of Liege, Liege, Belgium, for research to find lytic enzymes of microbial origin (5 years, 1964-1969); to the University of Lodz, Lodz, Poland, for research on the fermentative production of itatartaric acid (5 years, 1963-1968); to the University of Tokyo, Tokyo, Japan, for research on the fermentative production of D-tartaric acid (5 years, 1964-1969) and of mevalonic acid (3 years, 1965-1968); to the Institute of Biological Chemistry, University of Rome, Rome, Italy, for studies on the preparation and characterization of dextran derivatives (5 years, 1961-1966); to the National Sugar Institute, Kanpur, India, for research on isolation of natural polysaccharide gums (3 years, 1965-1968); and to the National Institute of Agronomic Investigations, Madrid, Spain, for study and collection of aerobic species of actinomycetes (4 years, 1965-1969).

PROGRAM OF STATE EXPERIMENT STATIONS

A total of 3.0 scientist man-years is devoted to this area of research.

PROGRESS -- USDA AND COOPERATIVE PROGRAMS

A. Chemical Composition, Physical Properties and Structure

1. Characterization of wheat gluten proteins. Separation of whole gliadin on Sephadex columns yielded fractions of differing molecular weights (the low-molecular-weight fraction contains α -, β -, and γ -gliadins). Sedimentation studies on whole gliadin and on these fractions, intact and after reduction and alkylation, indicated the following weight-average molecular weights respectively: whole gliadin, 43,000 and 26,000; high-molecular-weight fraction, 126,000 and 37,000; low-molecular-weight fraction, 27,000 and 22,000.

Studies on the composition of γ -gliadin resulted in its fractionation into three major components. Each had a distinct amino acid composition, electrophoretic mobility, and chromatographic behavior. Unlike γ_2 - and γ_3 -gliadin, γ_1 -gliadin was devoid of lysine and tryptophan and did not have aspartic acid as the N-terminal residue.

Glutenin was found to be more susceptible than gliadin to enzymic degradation. Behavior toward enzymes was consistent with the concept that disulfide bonds are intermolecular in glutenin and intramolecular in gliadin. The molecular weight of reduced alkylated γ -gliadin was found to be close to that of intact γ -gliadin in the same solvent system--a further indication of intramolecular disulfide bonding. Similar studies on reduced alkylated glutenin showed that its polypeptide subunits aggregated strongly even in powerful disaggregating solvents. The lowest apparent molecular weight for a subunit was 20,000. The reduction of disulfide bonds in glutenin could be controlled by use of less than stoichiometric amounts of mercaptoethanol and by variation of the pH of the reaction mixture. Further studies of optical rotatory dispersion indicated more α -helix in gliadin than in glutenin.

2. Desulfurization of wheat gluten proteins. Under the contract at Purdue University, further studies were conducted on the release of sulfur as sulfide at different levels of alkali and temperature, as well as in the presence of a reducing agent. The maximum yield of sulfide was around 15 percent of total sulfur at 25° C. but became about 75 percent at higher temperatures. In the presence of a reducing agent at 25° C., the liberation of sulfide from gliadin did not level off after 24 hours although it had reached a value of 75 percent of the cystine sulfur originally present.

Related research on cleavage of disulfide bonds in corn and wheat proteins is reported under Area 1, subheading A-3.

3. Chemistry of glycoprotein linkages. Under the grant to Marquette University on glycoproteins, study of a derivative of a glucosamine glycoside of serine showed that the glycoside carbohydrate moiety could

be removed by β -elimination with formation of derivatives of glucosamine and of α -aminoacrylic acid. The corresponding threonine derivative behaved similarly.

4. Microscopic and ultrastructure of wheat grain. Research on microscopic and ultrastructure of wheat grain and on changes induced therein by various treatments is relevant to industrial utilization of wheat. Results are reported under Area 5, subheading A-1.

5. NMR studies. Nuclear magnetic resonance techniques are employed in studies relevant to industrial utilization of wheat. Results are reported under Area 1, subheading A-4.

B. Chemical and Physical Investigations to Improve Products

1. Chemical modification of wheat gluten. Reduced whole gluten was chemically modified by graft copolymerization with methyl acrylate in dimethyl sulfoxide in the presence of sodium. The product was more soluble in organic solvents than the reduced gluten, but no longer dissolved in acid solutions. Studies on model compounds indicated that SH, NH₂, and CONH₂ groups reacted at about the same rate in graft copolymerizations of this type. Several S-alkyl cysteine derivatives were prepared as model compounds for study in connection with chemical modification of gluten.

Cyanoethylation of the amino group of tyrosine did not affect the pK of the phenolic group. This observation provided the basis for a novel differential potentiometric titration procedure for determination of amino and phenolic groups in aminophenols and in proteins. Terminally N-dicyanoethylated lysine was partially decarboxylated under conditions used for protein hydrolysis.

2. Polypeptide derivatives. Contract research at IIT Research Institute showed that sulfated polypeptides (obtained by hydrolysis of gluten) were less effective surfactants than some commercially available products. Polyether derivatives were prepared by reacting ethylene oxide with hydrolyzed gluten. By polymerization of acrylate monomers in the presence of these polyethers, products showing promise as adhesives were obtained. These polyethers also acted as plasticizers for films of some synthetic resins.

3. Chemical modification of wheat flour. Wheat flour reacted with ethylenimine (1.5 to 3 g./100 g. flour) imparted more strength to paper than did a commercial cationic sorghum flour, but slightly less than cationic corn starch.

4. Studies on wheat starch. Chemical and physical investigations on wheat starch are integrated with similar research on corn starch reported under Area 1, subheading B.

C. Microbiology and Fermentation

Research on microbiological and fermentative processes for converting wheat starch or flour to industrial products is integrated with similar studies on corn. Results are reported under Area 1, subheading C.

D. Technology - Process and Product Development

1. Milling and fractionation of wheat. Research on milling and fractionation of wheat is directed to production of improved products for industrial food and feed uses. Results are reported under Area 5, subheading C-1.

2. Acid-modified flour (AMF). In contract research at Iowa State University on fluidization of wheat flour, studies on HCl sorption rate were extended to fluidized beds where the rate was about 0.5 g./kg./min. from dilute concentrations of HCl in air. Ammonia was absorbed at about half the rate found for HCl. The specific heat of dry flour was determined to be 0.31 cal./g./°C. at 40-50°. At 12 percent moisture the value was 0.39. A continuous system for preparing AMF in a fluidized bed reactor was successfully operated. Products from soft white and hard red wheat flours had paste viscosities similar to those of products prepared conventionally at the Northern Division. This work essentially completes the activities specified in the contract.

Under a research contract, the Brown Company is evaluating AMF on a semi-commercial scale as a size for paper. Strength properties of paper sized with AMF were similar to those of paper treated with the reference size (a commercial hypochlorite-oxidized starch). During the sizing operations, which lasted about 2 hours, protein content doubled in the AMF pastes, but no interference with the operations or with properties of the paper produced was observed. Runs of longer duration are planned to determine if protein buildup reaches equilibrium or continues to increase.

3. Wheat-derived xanthates and xanthides in paper. In contract research at Battelle Memorial Institute, bran xanthate (D.S. 0.092) at the 10 percent level of addition increased strength properties of handsheets as follows: burst, 55 percent; dry tensile, 15 percent; and wet tensile, 1,100 percent. However, about 12 times the theoretical amounts of sodium hypochlorite was necessary to effect crosslinking of xanthate to xanthide. The cause for this excessive oxidant requirement is being sought. Gluten xanthate was ineffective as an additive.

4. Other developmental research. Development of products and processes involving wheat starch is integrated with related work on corn starch. Results are given under Area 1, subheading D.

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GRAIN SORGHUM UTILIZATION - FOOD
Northern Utilization Research and Development Division, ARS

Problem. An estimated 6 million bushels of grain sorghum are utilized annually in products for human consumption. This sorghum includes some special varieties such as white and waxy sorghums. Sorghum starch and derived glucose and glucose sirup are used in foods, and sorghum grits are used in fermented beverages. Although this outlet is at present quite limited, the growing importance of grain sorghum as a cash crop in the Southwest indicates that opportunities for increasing food use of sorghum should not be overlooked. Since grain sorghum is a staple food in many parts of Asia and Africa, a further consideration is the development of food products that could contribute to alleviation of dietary deficiencies in many developing countries.

To achieve the objective, more information on the composition of grain sorghum is needed. For example, some varieties contain pigments that can discolor milled products and that may contribute to undesired flavors. Questions have been raised concerning the digestibility and nutritive value of sorghum protein that reveal the need for better data on amino acid composition and on minor constituents.

Milling innovations, such as tangential abrasion, make possible conversion of about 20 percent of the sorghum kernel to a flour containing 25 percent protein. This and other possible approaches to new food products should be evaluated.

It has recently been discovered that certain oilseeds and cereal grains, including sorghum, are subject to infection by molds that can produce toxic products. To provide safe food products, as well as to minimize economic losses, research is needed on the detection of these toxins; on their quantitative analytical determination; and on development of processing techniques for their detoxification or removal from sorghum.

USDA AND COOPERATIVE PROGRAMS

The Department conducts a continuing, long-range program of research involving analytical, organic, and physical chemists, biochemists, microbiologists, systematic biologists, and chemical engineers engaged in basic, applied, and developmental studies pertinent to utilization of grain sorghum in food.

The Federal scientific effort for research on food utilization of grain sorghum totals .7 scientist man-year. Of this number, .3 is devoted to chemical composition and physical properties and .4 to microbiology and toxicology.

Research on chemical composition and physical properties involves a contract with Kansas State University, Manhattan, Kansas, for investigations on the

composition, processing, and feeding value of hybrid grain sorghum. A portion of this effort (.3 scientist man-year) is allocated to research on food uses of grain sorghum.

Research at Peoria, Illinois, on microbiology and toxicology (.3 scientist man-year) is devoted to studies on the production of mycotoxins by Aspergillus flavus and other molds. The work also includes a survey of the incidence of aflatoxin in commercial samples of various grains. A research contract in effect with the Agricultural Experiment Station, South Dakota State University, Brookings, South Dakota, provides for a survey of various species of Aspergilli to find and identify those producing toxic metabolites. A portion of this effort (.1 scientist man-year) is allocated to research on food uses of grain sorghum.

PROGRAM OF STATE EXPERIMENT STATIONS

State stations did not report research in this area.

PROGRESS -- USDA AND COOPERATIVE PROGRAMS

A. Chemical Composition and Physical Properties

1. Nutritional quality of grain sorghum. Contract studies on composition, processing, and feeding value of hybrid grain sorghums are important to food utilization of sorghum. Results are reported under Area 9, subheading A-1.

B. Microbiology and Toxicology

1. Aflatoxin investigations. Studies on toxins produced by molds are important to utilization of grain sorghum in foods. Results are reported under Area 3, subheading B-2.

PUBLICATIONS -- USDA AND COOPERATIVE PROGRAMS

None.

RELATED PUBLICATIONS OF STATE EXPERIMENT STATIONS

None.

GRAIN SORGHUM UTILIZATION - FEED

Northern Utilization Research and Development Division, ARS

Problem. The principal domestic use of grain sorghum produced in the U. S. is as feed for animals. The record 655-million-bushel crop in 1965 reveals the growing importance of this grain. About 80 percent of the crop is grown in Texas, Kansas, and Nebraska.

Problems are encountered in the use of grain sorghum in feeds which, if solved, could increase utilization and economic value of this crop to farmers and to the feed industry. The major need is for more and better information on the protein content and amino acid composition of various varieties of grain sorghum as related to biological feeding value. Minor constituents having physiological activity also require more adequate study. For example, certain phenolic pigments may impart bitterness and thereby reduce palatability. Carotenoid pigments, which in part are precursors for Vitamin A, are valuable in poultry rations for imparting yellow color to egg yolks and to the skin of fryers and broilers. In addition to such compositional studies, processing investigations are needed to provide ways for preserving desired and removing undesired components. Sorghum is included in the group of cereal grains and oilseeds recently recognized to be subject to infection by molds capable of producing toxic products. To provide safe feed products and to minimize economic losses, research is needed on the detection of these toxins; on their quantitative analytical determination; and on development of processing techniques for their detoxification or removal from grain sorghum.

USDA AND COOPERATIVE PROGRAMS

The Department conducts a continuing, long-range program of research involving analytical, organic, and physical chemists, biochemists, microbiologists, systematic biologists, and chemical engineers engaged in basic, applied, and developmental studies pertinent to utilization of grain sorghum in feed.

The Federal scientific effort for research on utilization of grain sorghum in feed totals 1.2 scientist man-years, of which .7 is devoted to chemical composition and physical properties and .5 to microbiology and toxicology.

Research at Peoria, Illinois, on chemical composition and physical properties involved study of carotenoid pigments of grain sorghum and has been completed. A research contract is in effect with Kansas State University, Manhattan, Kansas, for investigations on the composition, processing, and feeding value of hybrid grain sorghums. A portion of this effort (.7 scientist man-year) is allocated to research on feed uses of grain sorghum.

Research at Peoria, Illinois, on microbiology and toxicology (.3 scientist man-year) is concerned with studies on the production of mycotoxins by

Aspergillus flavus and related molds. Research contracts (.2 scientist man-year*) are in effect with A. D. Little, Inc., Cambridge, Massachusetts, for studies on stabilization of fermentative β -carotene; with Consolidated Laboratories, Inc., Chicago Heights, Illinois, for research on the use of antimetabolites to facilitate selection of higher yielding strains of microorganisms producing β -carotene; and with the Agricultural Experiment Station, South Dakota State University, Brookings, South Dakota, for survey of the genus Aspergillus to find and identify species producing toxic metabolites.

The Department also sponsors research in this area conducted under grants of PL 480 funds. Research on chemical composition and physical properties involves a grant to the Indian Institute of Science, Bangalore, India, for research on separation of grain sorghum proteins (5 years, 1963-1968).

Research on microbiology and toxicology involves a grant to the Agricultural University, Poznan, Poland, for studies to increase the yield of β -carotene produced by fermentation of cereal grains. Effort on this project is prorated among corn, wheat, and sorghum. During the reporting period, research was completed at the "Giuliana Ronzoni" Scientific Institute of Chemistry and Biochemistry, Milan, Italy, on production of Vitamin B₁₃ and at the National Institute for Agronomic Research, Paris, France, on mutation of yeasts for improved feeds.

PROGRAM OF STATE EXPERIMENT STATIONS

A total of 3.0 scientist man-years is devoted to this area of research.

PROGRESS -- USDA AND COOPERATIVE PROGRAMS

A. Chemical Composition and Physical Properties

1. Nutritional quality of grain sorghum. Preliminary results of contract research at Kansas State University showed that as protein level increases within a sorghum hybrid, all amino acids increase on an absolute weight basis. The relative amount of lysine, histidine, arginine, threonine, and glycine decreased, however, whereas that of glutamic acid, proline, alanine, and leucine increased. Apparently because of extremely favorable growing conditions in 1965, sorghum hybrids expected to exhibit the various levels of protein consistently observed in 1961, 1962, and 1963, instead exhibited essentially uniform protein levels.

2. Studies on sorghum proteins. At the Indian Institute of Science, qualitative differences were observed among disc electrophoresis patterns in polyacrylamide gels of protein fractions extracted with water, 1 percent NaCl, and 60 percent alcohol at 60° C. from three Indian varieties of sorghum. Fractions are being further characterized by gel electrophoresis after

*Work covers more than one commodity; only effort allocated to grain sorghum is included in total.

sulfide bond modification by reduction or by oxidation. One component of the alcohol-soluble protein fraction appears to contain no cystine. Experiments are in progress on the estimation of lysine present in protein hydrolyzates of about 40 varieties of sorghum seed of world-wide origin. This research is being conducted under a PL 480 grant.

B. Microbiology and Toxicology

1. Microbial carotenoids. Research on fermentative conversion of cereal grains to carotenoid-rich additives for feeds is applicable to grain sorghum. Results are reported under Area 3, subheading B-1.

2. Aflatoxin investigations. Studies on toxins produced by molds are important to utilization of grain sorghum in feeds. Results are reported under Area 3, subheading B-2.

3. Vitamin B₁₃. Studies on Vitamin B₁₃ are pertinent to utilization of grain sorghum in feeds. Results are reported under Area 3, subheading B-3 (PL 480 research).

4. Improved feeds by mutation of yeasts. Studies on mutant yeasts capable of producing high yields of sulfur-containing amino acids are relevant to feed utilization of grain sorghum. Results are reported under Area 3, subheading B-4 (PL 480 research).

PUBLICATIONS -- USDA AND COOPERATIVE PROGRAMS

None.

RELATED PUBLICATIONS OF STATE EXPERIMENT STATIONS

Chemical Composition and Physical Properties

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GRAIN SORGHUM UTILIZATION - INDUSTRIAL PRODUCTS
Northern Utilization Research and Development Division, ARS

Problem. The growing importance of grain sorghum as a competitive crop is revealed by the record 655-million-bushel crop in 1965. About 80 percent of the grain sorghum crop is grown in Texas, Kansas, and Nebraska. Sorghum starch and flour find industrial usage where freight transportation advantages exist. Currently an estimated 5 million bushels of sorghum are milled for products consumed mainly by the paper and gypsum board industries. To maintain this market against competition from synthetics and to take advantage of opportunities that exist in economically favorable geographic areas for increased industrial utilization of sorghum, technology suited to the specific characteristics of this grain and its milled products must be developed.

Research on sorghum starch is integrated with that on corn starch. A more detailed discussion of promising industrial outlets and of the pertinent research required is given under Area 1, Corn Utilization - Industrial Products. Because grain sorghum has a round kernel in contrast to the odd-shaped kernel of corn, it lends itself to dry milling innovations not possible with corn. Milling improvements, together with the possible advantages of air classification of sorghum flour, offer prospects for increasing industrial utilization by making processing economics more attractive and by providing products with superior properties.

USDA AND COOPERATIVE PROGRAMS

The Department conducts a continuing, long-range program of research involving analytical, organic, and physical chemists, biochemists, microbiologists, systematic biologists, and chemical engineers engaged in basic, applied, and developmental studies on the composition of grain sorghum, on characterization and properties of the components, and on their chemical and microbiological conversion to useful industrial products.

The Federal scientific effort for research on industrial utilization of grain sorghum totals 9.1 scientist man-years. Of this number, .9 is devoted to chemical composition, physical properties and structure; 3.3 to chemical and physical investigations to improve products; 3.0 to microbiology and fermentation; and 1.9 to technology - process and product development.

Research at Peoria, Illinois, on chemical composition, physical properties and structure (.2 scientist man-year) involves study of applications of nuclear magnetic resonance spectroscopy to grain components and is integrated with related research on corn.

A research contract is in effect with Indiana University Foundation, Bloomington, Indiana, for studies on the isolation and characterization of

phenolic pigments of grain sorghum (.6 scientist man-year). A grant (.1 scientist man-year*) to Iowa State University, Ames, Iowa, provides for basic research on heat, mass, and momentum transport of cereal starches and flours.

Research at Peoria, Illinois, on chemical and physical investigations to improve products (2.4 scientist man-years) is integrated with research on corn starch and is directed to wide-ranging study of the chemical reactions of starch with the objective of discovering new chemical products and processes having potential for industrial use. During the year, one phase of this work involving study of possible means for preparing amino acid derivatives of starch was completed and replaced by research on synthesis of halogen derivatives of starch. Research contracts (.4 scientist man-year*) are in effect with the Arizona Agricultural Experiment Station, University of Arizona, Tucson, Arizona, for basic studies on the reaction of acetylene with methyl glucoside; with The Johns Hopkins University, Baltimore, Maryland, for basic research on the reactions of starch in fluid dynamic media; with University of Pittsburgh, Pittsburgh, Pennsylvania, for studies on dielectric activation of starch; with Southern Illinois University, Carbondale, Illinois, for investigations on synthesis of maltooligosaccharides; with Stanford Research Institute, Menlo Park, California, for research on graft copolymers of cereal starches with vinyl-type monomers; and with the Institute of Paper Chemistry, Appleton, Wisconsin, for investigation of physical chemical factors affecting retention and effectiveness of starch xanthates and xanthides in paper. Contract research was completed by Ohio State University, Columbus, Ohio, on synthesis of amino derivatives of starch and by the University of Arizona Agricultural Experiment Station, Tucson, Arizona, on the reaction of starch with mercaptans. Grants (.5 scientist man-year*) have been made to Ohio State University Research Foundation, Columbus, Ohio, for basic research on the reaction of vinyl ethers with carbohydrates; to Ohio State University, Columbus, Ohio, for basic investigations of unsaturated and sulfur-containing carbohydrates and of the amination of starch; to Purdue Research Foundation, Lafayette, Indiana, for studies on sugars containing carbon-bound nitrogen, phosphorus and sulfur; and to the University of Arizona, Tucson, Arizona, for basic research on the reaction of starch with diepoxides.

Research on microbiology and fermentation conducted at Peoria, Illinois (2.6 scientist man-years), includes studies on the use of microorganisms to convert cereal-based media to industrially useful products such as chemicals, enzymes, polymers, and biological insecticides. This research is integrated with similar studies based on corn. A large collection of pure cultures of industrially and agriculturally important microorganisms is maintained. The Pioneering Laboratory for Microbiological Chemistry

*Work covers more than one commodity; only effort allocated to grain sorghum is included in total.

conducts research on microbiological reactions and products. Investigations on biological insecticides for Japanese beetle and on other insect control agents is cooperative with Entomology Research Division and Plant Pest Control Division. Research on plant antibiotics involves cooperation with Crops Research Division. Research contracts (.1 scientist man-year*) are in effect at Michigan State University, East Lansing, Michigan, for basic research on enzyme activity in sporulation; at Kansas State University, Manhattan, Kansas, for investigation of stabilization of vegetative cells of the pathogenic organisms; at the University of Minnesota, St. Paul, Minnesota, for fundamental studies on the transfer of genetic determinants of sporulation from one microorganism to another; at Baylor University, Houston, Texas, for investigation of morphological changes involved in sporulation; and at the American Type Culture Collection, Rockville, Maryland, for studies on preservation of certain microorganisms for which lyophilization is ineffective. Contract research at the University of Illinois, Urbana, Illinois, for research on the applicability of a sporulation factor produced by bacteria to Japanese beetle pathogens has been completed. Grants (.3 scientist man-year*) have been made to Cornell University, Ithaca, New York, for fundamental studies on biphasic fermentation; to the Nebraska Agricultural Experiment Station, University of Nebraska, Lincoln, Nebraska, for investigations on the nature of amylase enzymes; to Kansas State University, Manhattan, Kansas, for investigations on separation of enzymes and proteins by disc electrophoresis; to Iowa State University, Ames, Iowa, for investigation on bacterial amylases and their action patterns; to the University of Wisconsin, Madison, Wisconsin, for studies on the fine structure of polysaccharide B-1973; and to the University of Arkansas, Fayetteville, Arkansas, for investigation of the mechanism of enzymatic hydrolysis of starch.

Research conducted at Peoria, Illinois, on technology - process and product development (1.2 scientist man-years) is concerned with detailed study and evaluation of starch derivatives having definite potential for industrial utilization and of processes for making them. The work is integrated with similar studies on corn starch derivatives. Research contracts (.7 scientist man-year*) are in effect with Stanford Research Institute, Menlo Park, California, for process development of selected starch graft copolymers; with Battelle Memorial Institute, Columbus, Ohio, for developmental research on starch and other cereal grain xanthides and for studies on starch derivatives for use as colloids in water-emulsion paints; with Western Michigan University, Kalamazoo, Michigan, for evaluation of modified cyanoethylated starches for applications in paper; with Archer Daniels Midland Company, Minneapolis, Minnesota, for investigations on the use of starch glycosides in coatings and plastics; and with University of Akron, Akron, Ohio, for evaluation of starch and starch derivatives as reinforcing agents for

*Work covers more than one commodity; only effort allocated to grain sorghum is included in total.

natural and synthetic rubber. During the year, contract research on evaluation of starch polyol urethane foams was completed by Archer Daniels Midland Company, Minneapolis, Minnesota.

The Department also sponsors research on cereal starches conducted by foreign institutions under grants of PL 480 funds.* Research on chemical composition, physical properties and structure involves grants to the University of London, London, England, for research on debranching enzymes and their use in studying the fine structure of starch components (5 years, 1963-1968); to the University of Osaka Prefecture, Sakai, Japan, for development of an analytical method for carbonyl groups in carbohydrates (4 years, 1964-1968); and to "Giuliana Ronzoni" Scientific Institute for Chemistry and Biochemistry, Milan, Italy, for research on glucopyranose rings in starches and dextrans (5 years, 1962-1967).

Research on chemical and physical investigations to improve products involves grants to Hebrew University, Jerusalem, Israel, for studies on starch vinyl and epoxide graft copolymers (4 years, 1963-1967); National Institute of Technology, Rio de Janeiro, Brazil, for research on phosphorus- and sulfur-containing cationic starches (5 years, 1962-1967); Ahmedabad Textile Industry's Research Association, Ahmedabad, India, for research on starch-gum copolymers prepared by codextrinization (5 years, 1963-1968), and for studies on preparation and characterization of hydroxyethyl ethers of cereal starches (5 years, 1965-1970); Academy of Sciences and Chemical Institute "Boris Kidric", Ljubljana, Yugoslavia, for studies on modification of starch by moisture and temperature treatments (5 years, 1964-1969); Plastics Research Institute TNO, Delft, The Netherlands, for research on preparation of metal alkoxides of starch for use as intermediates in synthesis (5 years, 1964-1969); and University of Edinburgh, Edinburgh, Scotland, for studies on the mechanism and structural changes involved in thermal, acid, and alkaline degradation of starches (5 years, 1964-1969); and to the Institute of Fibres and Forest Products Research, Jerusalem, Israel, for studies on the mechanism and products of mild oxidation of starch (5 years, 1963-1968). During the year, research was completed on fatty chemical derivatives of starch dextrans at the Institute of Industrial Chemistry, Bologna, Italy, and on changes induced in starch by gamma-irradiation at the National Institute of Agronomic Research, Paris, France.

Research on microbiology and fermentation involves grants to the University of Milan, Milan, Italy, for basic studies on the metabolic pathway to 5-ketogluconic acid in Acetobacter species (5 years, 1961-1966); University of Allahabad, Allahabad, India, for collection of new Mucorales species (5 years, 1961-1966), and studies on survival of lyophilized microorganisms (5 years, 1962-1967); University of Newcastle upon Tyne (formerly University of Durham), Newcastle upon Tyne, England, for investigations of sugar phosphate derivatives in molds (5 years, 1962-1967); Central Drug Research Institute, Lucknow, India, for studies on aerobic actinomycetes in India to

*Effort prorated among corn, wheat, and grain sorghum.

find new accessions for the ARS Culture Collection (5 years, 1965-1970); to the University of Liege, Liege, Belgium, for research to find lytic enzymes of microbial origin (5 years, 1964-1969); to the University of Lodz, Lodz, Poland, for research on the fermentative production of itatartaric acid (5 years, 1963-1968); University of Tokyo, Tokyo, Japan, for research on the fermentative production of D-tartaric acid (5 years, 1964-1969) and of mevalonic acid (3 years, 1965-1968); to the Institute of Biological Chemistry, University of Rome, Rome, Italy, for studies on the preparation and characterization of dextran derivatives (5 years, 1961-1966); to the National Sugar Institute, Kanpur, India, for research on isolation of natural polysaccharide gums (3 years, 1965-1968); and to the National Institute of Agronomic Investigations, Madrid, Spain, for study and collection of aerobic species of actinomycetes (4 years, 1965-1969).

PROGRAM OF STATE EXPERIMENT STATIONS

State stations did not report research in this area.

PROGRESS -- USDA AND COOPERATIVE PROGRAMS

A. Chemical Composition, Physical Properties and Structure

1. Phenolic pigments of grain sorghum. In contract research at Indiana University, methanol extraction was found to remove pigments from dewaxed sorghum pearlins. Six or more leucoanthocyanidins or leucoanthocyanins were detected in the methanol extract of dewaxed sorghum pearlins. Also, treatment of this methanol extract with lead acetate yielded five or more substances of the coumarin or cinnamate type. Methods for characterizing phenolic pigments in the extracts are being investigated.

2. NMR studies. Nuclear magnetic resonance techniques are employed in studies relevant to industrial utilization of grain sorghum. Results are reported under Area 1, subheading A-4.

B. Chemical and Physical Investigations to Improve Products

C. Microbiology and Fermentation

D. Technology - Process and Product Development

Research in these categories is integrated with similar investigations on corn starch. Results are reported in Area 1, subheadings B, C, and D.

PUBLICATIONS -- USDA AND COOPERATIVE PROGRAMS

Chemical Composition, Physical Properties and Structure

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Technology - Process and Product Development

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RELATED PUBLICATIONS OF STATE EXPERIMENT STATIONS

None.

FORAGE UTILIZATION - FEED

Northern Utilization Research and Development Division, ARS

Problem. Tall fescue grass is grown extensively in the Southeast, in the Intermountain States, and in the Pacific Northwest as a forage crop for cattle and other domestic animals. It has excellent agronomic characteristics, producing well on marginal land and remaining green during cool weather when other grasses are dormant. The quality of staying green in the winter is a prime factor in its acceptance. Thirty-five to fifty million acres of fescue are grown for forage use in the Southeastern part of the United States alone.

Cattle grazing on pasture that is predominately tall fescue sometimes develop a disease known as "fescue foot." In severe attacks the animal first becomes lame. The peripheral portion of one or more limbs then develops necrosis, and sloughing of the hooves may occur. Occasionally the tail and ears may be affected. Animals become emaciated and frequently die. The disorder is more apt to occur during cool weather than during the summer months. However, even when conditions are not such as to produce the more dramatic symptoms, cattle sometimes perform poorly on fescue forage, a result which may be attributed to subclinical toxicity.

Pastures may become toxic after several years of freedom from toxicity. Serious outbreaks of fescue toxicity occurred during the winter 1963-64 in parts of Kentucky, Illinois, Missouri, Kansas, and Arkansas. Thousands of head were involved, with morbidity ranging from 1 percent to 99 percent of the herds. For example, 42 of 72 head of cattle became lame after 8 days on one pasture in Missouri. In these outbreaks, the toxic pastures were soil bank lands having long grass that was pastured after the advent of cold weather.

Research to determine the cause of toxicity in fescue and to identify the toxic substance(s) is needed as a basic step in developing a solution to the problem of toxic fescue.

USDA AND COOPERATIVE PROGRAMS

At the Northern Division, Peoria, Illinois, the Department has a program of limited scope that involves one organic chemist engaged in research to isolate and identify the toxic component(s) of tall fescue grass responsible for a cattle disease known as "fescue foot." This research is cooperative with the Kentucky State Experiment Station, which furnishes toxic and non-toxic fescue grass for chemical study and conducts bioassays of fractions and components isolated from fescue at the Northern Division. Liaison is maintained with the fescue breeding program of the Field Crops Research Branch, ARS, through the Agronomy Department of the University of Kentucky and with the Department's Pharmacology Laboratory at the Western Division.

The major part of the Department's research program on forages is maintained at the Western Utilization Research and Development Division, Albany, California.

The Federal program at Peoria, Illinois, totals 1.3 scientist man-years, all of which is devoted to microbiology and toxicology.

PROGRAM OF STATE EXPERIMENT STATIONS

A total of 10.0 scientist man-years is devoted to this area of research.

PROGRESS -- USDA AND COOPERATIVE PROGRAMS

A. Microbiology and Toxicology

1. Fescue toxicity. For the first time, toxicity to the bovine has been demonstrated for a metabolite of a mold isolated from toxic fescue and cultured on non-toxic fescue. Fusarium nivale was grown on non-toxic hay enriched with glucose and peptone. The concentrated 80-percent alcohol extract from this culture gave a positive result in a small animal assay and was toxic to the cow. A crystalline compound of the butenolide class has been isolated from the extracts of F. nivale cultures and found to give a positive test in the small animal assay. The structure of the compound has been determined. Isolation of this irritant, belonging to a class noted for physiological activity, from a mold originally found on fescue, justifies further intensive study even though this compound per se may not be the cause of fescue foot. The work involved cooperation with the Western Division and the Kentucky Agricultural Experiment Station.

PUBLICATIONS -- USDA AND COOPERATIVE PROGRAMS

None.

RELATED PUBLICATIONS OF STATE EXPERIMENT STATIONS

Chemical Composition, Physical Properties and Structure

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FORAGE UTILIZATION - FEED

Western Utilization Research and Development Division, ARS

Problem. The demand for livestock in the United States will increase 45% by 1975. Since forage crops constitute the major feedstuff for ruminant animals, the demand for forages will increase accordingly. In addition, there is an increasing demand for processed forages in European and Asiatic markets. Fresh forage crops are the richest natural source of many nutrients for farm animals. Forages, however, are preserved so inefficiently by hay-making and ensiling that 10 to 50% of the dry weight and much larger fractions of the most valuable nutrients are lost before the animals eat them. Dehydration is now the only practical means of producing products of high nutritional value in a form usable in manufactured feeds and supplements. Poultry and swine producers are aware of the value of dehydrated forage, but restrict their use of it because of its high fiber and growth-inhibitor content. There is evidence that certain unidentified growth factors are lost, at least partially, during the dehydration process as presently carried out. The livestock breeder needs forage products tailored to specific animals, and the forage producer must adapt to his needs to sell.

Basic and applied utilization research are necessary to produce: (1) high-protein, low-fiber feeds rich in unidentified growth factors designed for use by non-ruminant animals; (2) fiber products which have been cheaply treated to make them easily digestible for ruminants; (3) growth-stimulating supplements for ruminants, derived from the biologically active fiber-digestion factors and growth-promoting factors in forage. New products should be adaptable to mechanical feeding. Improved uses will encourage farmers to put high-value land into forage crops.

USDA AND COOPERATIVE PROGRAM

Current research in the Western Utilization Research and Development Division includes both basic and applied studies on all forages used or potentially usable for off-the-farm processing. The research is conducted at the Division headquarters at Albany, California; under contracts at Berkeley, California, Lincoln, Nebraska, and Athens, Georgia; and Under P.L. 480 grant programs in Scotland and Italy. Basic compositional studies deal with the potent estrogen, coumestrol (discovered by Department scientists), and other phenolic compounds present in forage legumes. The value of coumestrol-rich alfalfa as a growth stimulant for ruminants is being studied in cooperation with Oregon State University. The mechanical separation of leaf from stem of alfalfa is being studied with financial support from the Department of Agriculture and Inspection of the State of Nebraska and the cooperation of several experiment stations and commercial processors and users of forages. Also under study are biologically active forage constituents (such as the chick-growth-promoting factor in forage juices and alfalfa saponins which depress chick growth), organic acids of alfalfa, non-protein nitrogen compounds of alfalfa, and the mechanism of action of forage antioxidants.

Processing of forages by "wet" (juicing) and "dry" (turbomilling and air classification) methods is being investigated. The effects of dehydration conditions on losses of carotene and xanthophyll are being studied.

The Federal program of research in this area totals 7.4 scientist man-years, including one scientist whose salary is provided by the Department of Agriculture and Inspection, State of Nebraska, and contract research equivalent to 2.0 scientist man-years per year. Of this number 4.5 are assigned to chemical composition and physical properties and 2.9 to technology--process and product development. In addition the Division sponsors, under P.L. 480, two research projects on forage composition.

PROGRAM OF STATE EXPERIMENT STATIONS

A total of 10 scientist man-years is devoted to this area of research.

PROGRESS -- USDA AND COOPERATIVE PROGRAMS

A. Chemical Composition and Physical Properties

1. Forage Composition. Because of their association with biological activity, phenolic compounds of alfalfa and other forages are being isolated and identified. Two compounds from ladino clover, in addition to those reported earlier, have been characterized as 7-hydroxycoumarin and 7,7'-dihydroxy-6,8'-bicoumarinyl. The structure of two previously unknown compounds from alfalfa has been confirmed by unequivocal synthesis as 7-hydroxy-11,12-dimethoxycoumestan and 7,12-dihydroxy-11-methoxycoumestan. A series of 20 coumestans isomeric with coumestrol and its mono- and dimethoxy derivatives have been synthesized for chemical, physical, and biological comparison with coumestrol.

Several previously unidentified acids of alfalfa have been tentatively identified as monosodium and monoethyl derivatives of dicarboxylic acid. During the study of previously known constituents by means of new analytical methods, new unidentified acids have been detected. Analyses of holocellulose and lignin by several procedures on a series of alfalfa samples have been completed. We will determine which of these procedures correlates best with metabolizable energy value for chicks.

Research on a method for analyzing xanthophyll has led to a new cold saponification step which eliminates the chlorophyll error in methods previously available. The new procedure is simplified yet accurate compared with other methods. It has been adopted for a collaborative evaluation by industry and should give the feed formulator confidence in employing alfalfa meal as the source of xanthophyll pigmenters in poultry feed. In studies of dehydration, the effects of moisture of meal and inlet and outlet temperatures on the xanthophyll content and extent of carotene isomerization are being examined. As much as 50% of the xanthophyll content of fresh alfalfa may be destroyed by improper dehydration procedures.

In cooperative work with the Nutrition Department at the University of California, evidence has been found for a growth factor in dehydrated alfalfa. By adding 10% of dehydrated alfalfa to the diet of guinea pigs, improvement in growth of up to 50 g. over a 3-week assay period was obtained.

The factor(s) appears to be distinct from all known vitamins, minerals, amino acids, and other nutrients and appears to be organic in nature, since it is not found in the ash of alfalfa. Studies are now in progress to define the properties and nature of this unidentified factor(s).

2. Interactions of Forage Antioxidants. Pigments, vitamin E, lipids and related substances provide much of the biological value of alfalfa, but they are subject to oxidative deterioration. Contract research on chemical changes of alfalfa lipids is being conducted at the University of California at Berkeley to provide basic information that could lead to stabilization of alfalfa and other forage products. The lipid composition of fresh alfalfa and changes that occurred immediately after cutting were determined. Beta-carotene content dropped rapidly immediately after cutting. Linolenic acid, the major fatty acid in alfalfa lipids, did not decrease until after the first 24 hours but then rapidly decreased to less than 50% of its original value in 48 hours. Linolenic acid is converted to products that have been extremely difficult to extract from alfalfa. Some minor increases occurred in palmitic, stearic, and oleic acid, but other fatty acids did not change. Since about 90% of the fatty acid present in alfalfa galactolipids is linolenic acid, substantial changes must take place in the galactolipids as harvested alfalfa is field cured over a two- to several-day period. Monogalactolipids are more prone to change than digalactolipids. Galactolipids affect antioxidant activity, so experiments were initiated to determine the effect of antioxidants on the rate of disappearance of beta-carotene and linolenic acid in alfalfa. In addition, the natural antioxidants from alfalfa were tested on other oxidizable lipids. Monogalactolipid was effective as an antioxidant for menhaden oil but showed no antioxidant activity for squalene. Digalactolipid was not effective as an antioxidant. The monogalactolipid content of alfalfa is about twice the digalactolipid content.

Under a P.L. 480 grant, research at the Experiment Station for Practical Agriculture in Milan, Italy, is underway on non-tocopherol reducing substances in alfalfa that may be involved in the natural antioxidant activity of alfalfa products. Natural antioxidants exist in alfalfa with activity that is several times as great as the antioxidant activity of tocopherol. Methods for separation, identification, and assay were developed that reduced to a minimum the alteration and destruction of the compounds during the laboratory procedures. Qualitative recognition and semiquantitative evaluation of the substances were carried out. At least four reducing compounds, whose evolution was followed during the growth of the plant, were isolated. During regrowth of alfalfa after cutting, one of the compounds increased in large amounts during the first vegetative preflowering stage. Another compound, absent during the first week of vegetation, resolved into three distinct compounds, one of which predominated over the other to become four or

five times more abundant by the time of flowering. During the winter, no appreciable amounts of these reducing compounds could be identified, either in hibernating plants in the open field or in those vegetating in green-houses. These non-tocopherol reducing agents are probably formed during the plant's period of highest photosynthetic activity. Chromatographic RF value provided a tentative identification of one of the compounds.

3. Structure of Alfalfa Polysaccharides. Research is being conducted under P.L. 480 funds at Edinburgh University in Scotland to determine the nature of alfalfa polysaccharides and to investigate enzyme systems that may be helpful in structural analysis of the polysaccharides and components associated with them. The amylase activity of alfalfa seedling extracts is very similar to that of malt alpha amylase. Amylase can split large molecules made up of 6-carbon glucose. The extracts from alfalfa also show enzyme activity in hydrolyzing large compounds made of the 5-carbon sugar xylose. The main hemicellulosic component of alfalfa stems has been characterized as to its constituent carbohydrates. A similar component is present in the alfalfa leaf, and structural examination has been conducted in sufficient detail to establish its essential identity to the stem hemicellulose. Sufficient quantities of oligosaccharides and D-galacturonic acid were prepared for a study to be made of the enzyme activities that will hydrolyze these compounds.

B. Technology--Process and Product Development

1. Improved Feeds from Forages. Preliminary tests with high-estrogen forages or extracts prepared from them indicate that naturally occurring plant estrogens have growth-promoting effects on sheep. Four feeding trials with lambs fed varying amounts of alfalfa coumestrol were conducted at Oregon State University on a cooperative basis. Tests were made with crude alfalfa meals, acetone extracts of different coumestrol potencies, and with isolated coumestrol. Except with the isolated coumestrol, a trend toward positive growth response to elevated coumestrol levels was obtained with wether lambs but not with ewes. Estrogenic responses were observed in animals fed the higher levels of coumestrol. In sensory tests, lamb roasts from animals fed high coumestrol diets consistently scored higher in tenderness and juiciness.

A study was conducted cooperatively with the Crops Research Division of the Agricultural Research Service and Purdue University to relate coumestrol content of alfalfa to growing area, variety, cutting, harvest year, stage of growth, and disease. It was found that the variation in coumestrol content was essentially nongenetic. In alfalfa samples infected with common leaf spot, a definite increase in concentration of coumestrol was observed as the infection became more severe. The relative amount of the group of phenolics accompanying coumestrol to the amount of coumestrol present was roughly the same in lightly and heavily infected samples. Amounts in disease-free plants were indiscernible. It seems reasonable to assume that the build-up of at least eight other phenolic compounds in plants parallels the build-up of coumestrol during infection.

Mixed feeds for poultry require a yellow pigmentation source. The use of alfalfa in mixed feeds is based principally on its xanthophyll content. The pigmentation potency of various xanthophyll sources was determined by chick feeding experiments. Yellow corn meal, corn gluten meal, pelleted and reground alfalfa meal, lutein, and isolated carotenoid and xanthophyll extracts of alfalfa were compared. The extracts from alfalfa showed higher biological availability of yellow pigment for chicks than did alfalfa, itself, or corn gluten meal. Pure lutein, one of the xanthophylls from alfalfa showed greater pigmentation potency than did the extract which contained a mixture of xanthophylls. The mixed xanthophyll of dehydrated alfalfa meal was as well utilized by hens for egg yolk pigmentation as was that of the alfalfa extract.

2. Alfalfa Processing. A mobile processing unit was operated in Nebraska in cooperation with the State of Nebraska Department of Agriculture and Economic Development, Nebraska Farm Products, Inc., and the Kansas-Nebraska Natural Gas Company. Two cuttings from four plots of alfalfa harvested at 28, 32, 36 and 40-day cycles provided the raw material. Dehydrated alfalfa was separated into three fractions: leaf, stem, and fines. The separate materials will enable the processor to produce feed specifically for the different classes of livestock (e.g., mono-gastric animals and ruminants). Longer intervals between alfalfa cuttings allow a gross increase in yields per acre and strengthen stands. However, usual practice precludes the longer interval because much of the increased yield is in fibrous material as the stand matures. By use of the new leaf separation process, alfalfa meal of 25% to 30% protein grade is still available while yielding as much or more of the stem fraction suitable for feeding ruminants. One dehydrator plant now has several hundred tons of super grade alfalfa at about 27% protein available for market testing, and other operators are keenly interested. Improved control systems are being installed in our mobile processing unit. Tests are to be conducted at Dixon, California in cooperation with the Dixon Dryer Company in 1966. Further tests using commercial-scale equipment will be conducted in Nebraska this summer.

Contract research has been initiated at the University of Nebraska in Lincoln to evaluate, in mixed feeds, alfalfa products that have been processed to lessen the effects of high fiber content. Artificial rumen techniques are being used in preliminary screening studies. Wet ball milling of alfalfa stem increases the rate of cellulose digestibility and the total percentage digested. In one test, cellulose digested in 32 hours increased from 35% to 60% of the alfalfa sample. The State of Nebraska Department of Agriculture and Economic Development and the Nebraska Farm Products, Inc. of Cozad, Nebraska have cooperated in this project.

3. Products from Southeastern Grasses. Contract research is being conducted at the University of Georgia in Athens to study products that can be made by dehydrating Coastal Bermuda grass and pearl millet. Attention has been given to production of higher quality forage and to evaluation and development of

methods for carotenoid pigment analysis. Nineteen cuttings of Coastal Bermuda were made, including 8 cuttings at 3-week intervals, 6 at 4-week intervals, and 5 at 5-week intervals. With 3- to 4-week cutting cycles, good quality was maintained; it decreased slightly as the season progressed to hot weather but increased again as cooler weather started.

With millet, quality can be improved by varying the cutting cycles with the seasons. Millet grown in the Southeast was consistently higher than Coastal Bermuda in moisture content. Both grasses contain high levels of carotene and xanthophyll compared with standard grades of alfalfa meal. Carotenoid stability in dehydrated grasses during storage is similar to that of alfalfa meal and is similarly enhanced by use of the antioxidant ethoxyquin. This research is expected to lead to increases in production of high-quality dehydrated forage products that will be important because of their nearness to the large broiler-producing areas of Georgia and the Delmarva Peninsula. Such products will help regain feed markets lost in these areas because of the high fiber-to-nutrient ratio of alfalfa meal. The products should move into export markets as well.

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RICE UTILIZATION - FOOD

Southern Utilization Research and Development Division, ARS

Problem. The productive capacity of U. S. rice growers has increased faster than domestic and export consumption over the past decade, thus limiting the income potentially available from this major world food grain. Detailed knowledge of chemical composition and physical properties as related to processing is needed to guide milling, processing, and product development of U. S. rices so that they can better meet the quality and new product requirements needed for expanded markets. New and diverse food products that are economical to manufacture, convenient to prepare, and attractive in flavor and texture are needed to increase the total consumption of rice both domestically and abroad. Additional needs include the development of improved milling machinery and techniques, primarily to increase the yield of head rice; intensified research on deep milling to evaluate and utilize the protein flour and residual kernels produced by this technique; and research to provide greater flexibility in the industry by developing from either medium or long-grain rice new products that can be cooked to provide either discrete kernels or a gelatinous food.

USDA AND COOPERATIVE PROGRAM

The Department has a continuing long-term program at New Orleans, Louisiana, involving biochemists and analytical chemists engaged in research on the chemical composition and physical properties of rice and its products. Distribution of the chemical constituents of milled rice in consecutive layers of the kernel is being studied, with special emphasis on nutritionally important constituents such as proteins, amino acids, starch, lipids, vitamins, and minerals; and on the cooking and chemical characteristics of the kernels remaining after differential removal of these layers. Findings from this research will provide the necessary basis for evaluating the economic feasibility of using high-protein rice flours (layers removed by deep milling) for protein fortification of foods and as dietetic or other specialty foods. Close cooperation is maintained, under formal memoranda of understanding, with the Louisiana, Arkansas, and Texas Rice Experiment Stations, who supply rice samples of known variety and cultural history for the experimental studies. The Rice Inspection Service, Grain Division, AMS, New Orleans, Louisiana, cooperates by providing assistance in grading rice samples from the research investigations. Cooperation is also maintained with the Western Division.

Other research on chemical composition and physical properties has been initiated under a P. L. 480 grant to Kyoto University, Kyoto, Japan. Scientists at this institution will study the distribution of the major proteins of rice within subcellular particles and the distribution of these particles in the cellular structure of the rice kernel to obtain basic information needed to develop new and improved rice products and methods of producing them (project duration--3 yrs.).

The Federal in-house scientific effort devoted to research in this area totals 1.7 scientific man-years, all of which are presently applied to chemical composition and physical properties.

Under P. L. 480 research there is presently one grant, also on chemical composition and physical properties.

PROGRAM OF STATE EXPERIMENT STATIONS

A total of 3.0 scientific man-years is devoted to this area of research.

PROGRESS -- USDA AND COOPERATIVE PROGRAMS

A. Chemical Composition and Physical Properties

1. Investigation of the Distribution of Chemical Constituents in the Rice Kernel. The physical properties of high-protein rice flours, which were prepared by tangentially abrading successive layers from conventionally milled Bluebonnet-50 long-grain rice, were investigated. Determination of the hot paste viscometric properties and pasting characteristics of the eight successive flour layers (each representing about 6-7% by weight of the original milled rice) showed that none of the layers exhibited a peak or set-back when the rice paste was heated to and above the gelatinization temperature. In contrast, the hot paste viscosity of the original milled rice peaked at 1250 Brabender Units (BU). The residual kernels obtained after removal of each of the layers exhibited successive increases in peak viscosity--1450 to 1800 BU--from the first to the sixth layer. An unexplained decrease in peak viscosity--1660 to 1600 BU--was shown by the kernels remaining after the removal of the seventh and eighth layers. The viscometric properties of these high-protein rice flours are quite unique and different from those of any commercial rice flour, and must be taken into consideration in the development of new uses for these flours.

The chemical composition and viscometric properties of flours prepared from whole, second heads, and parboiled rice in a commercial Satake mill were investigated to determine feasibility of adapting commercial equipment to high-protein flour preparation. One and a quarter tons of Belle Patna rice (7.3% protein, 0.9% fat, 91.7% starch) was abrasively milled and five successive fractions separately collected for a total of 14.4% wt. removal. Flour:chits ratio (80-mesh) was approximately 60:40 except for 40:60 in fraction 1. Protein in flour progressively decreased from 15.1 to 14.5%; fat 12.8 to 1.7%; and starch increased 42.9 to 79.1% in successive fractions. Values of the chits were: 11.7 to 8.5% protein, 6.1 to 0.3% fat, and 65.5 to 89% starch. Brabender viscograms of the 5 flours indicated unusually low values for the pasting characteristics with a slight progressive increase from fractions 1 to 5. Residual kernels exhibited progressively higher values of pasting characteristics than did the initial feed. Chits were intermediate in values, tending to approach those of the residual

kernels. Similar analyses were conducted on high-protein flour prepared by removal of two successive fractions from Belle Patna second heads and from parboiled rice. In addition, two 200-lb. samples of flour were commercially prepared from head and second head Belle Patna for sample distribution. This evaluation of flours prepared with the Satake mill demonstrates that it may be possible to adapt commercial equipment to high-protein rice flour preparation. The development of the product can be a useful adjunct to the domestic rice milling and reprocessing industries in that it provides a whiter rice with improved cooking characteristics and a high-protein flour that may be useful in many domestic dietary products. Since the residual kernels are high in starch and low in protein and fat, they should also provide more uniform stock for industrial uses. (Sl 4-13).

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RICE UTILIZATION - FOOD

Western Utilization Research and Development Division, ARS

Problem. Limitations on U.S. rice acreage are being relaxed because of the increasing need for rice to export into food-short areas, particularly Southeast Asia. Polished rice is a relatively expensive source of nutrients because its protein, although high in quality, is only about 5% to 7% of the kernel, and it is almost devoid of vitamins. The need is for new and improved food products with better nutritive quality and produced at minimum increase in cost. Such products must be easy to prepare; have good texture, flavor, and appearance; and be economical to manufacture. Also needed are drastically improved milling methods to increase economic returns to growers and millers so that cost to consumers will not increase. Detailed knowledge of chemical composition and physical properties as related to processing is needed to guide the developments in milling, processing, and development of products that meet the growing export demand.

USDA AND COOPERATIVE PROGRAM

In the Western Utilization Research and Development Division, basic and applied research on rice is conducted at Albany, California. Basic studies are concentrating on rice proteins. Process research is underway on debranning of brown rice by lye-peeling, parboiling of brown and under-milled rice, new methods to produce quick-cooking forms having better flavor and texture, and conversion of high-protein flours into beverage products especially suitable for infant feeding overseas. Cooperative studies with industry and the University of California are conducted on improved methods for drying rice.

The Federal program of research in this area totals 2.4 professional man-years. Of this number 1.2 are assigned to chemical composition and physical properties and 1.2 to technology--process and product development.

PROGRAM OF STATE EXPERIMENT STATIONS

A total of 3 scientist man-years is devoted to this area of research.

PROGRESS -- USDA AND COOPERATIVE PROGRAMS

A. Chemical Composition and Physical Properties

1. Rice Proteins. Studies in the past few years have begun to supply knowledge on the fundamental makeup of rice proteins. We are continuing these studies in order to establish a firmer base on which to develop high-protein rice foods. Extracts of rice prolamines (proteins which are soluble in 60-80% alcohol) have been resolved into seven components, one of which is predominant and four very minor. All of these components

differ distinctly from the components of globulin proteins (proteins which are soluble in a salt solution). The prolamine constituents are being separated, in order to determine individual amino acid compositions.

Isolated rice globulins have been segregated into single components on the basis of solubility differences. One protein component comprising 40-45% of rice globulins has been partially characterized. It has a molecular weight of about 25,000 and is rich in arginine and glutamic acid; it contains considerable cystine and methionine and is almost devoid of lysine and histidine. The outer layers of rice endosperm (see discussion on high-protein rice flours, Paragraph B. 1. below) contain higher proportions of soluble proteins; a decreasing gradient in biological value of protein may exist as well as a decreasing gradient in concentration of protein.

B. Technology--Process and Product Development

1. Improved Rice Products. Abrasive milling of flour from surface layers of milled rice in commercial mills is possible with all types of rice. Flours with about double the protein content of the original kernels have been produced. Dry powders that are easily dispersible in cold water to form a milk-like beverage have been made from high-protein flours of both regular and parboiled rice by two processes, one involving partial hydrolysis of protein by pepsin and the other using amylase plus pepsin to hydrolyze the starch as well as the protein. Simple mixing of the dry solids with water produces beverage foods of milk-like consistency.

High-protein flour obtained by deep milling of parboiled rice is practically equivalent in protein efficiency value to similar flour obtained from white milled rice. This indicates that the parboiling process does not significantly reduce the nutritive quality of the rice protein. In addition to doubling the protein content, deep milling provides several-fold increases in the vitamins thiamine and riboflavin and in fats and minerals.

PUBLICATIONS - USDA AND COOPERATIVE PROGRAMS

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RELATED PUBLICATIONS OF STATE EXPERIMENT STATIONS

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Technology -- Process and Product Development

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NUTRITION AND CONSUMER USE RESEARCH

Consumer and Food Economics Research Division, ARS
Human Nutrition Research Division, ARS

Problem. The assortment and characteristics of food available to consumers change constantly with the adoption of new practices of production, processing, and marketing. Changing constantly also, as nutrition science advances, is our understanding of the nutritional needs of man and the manner in which these needs can best be met by food. To help meet the Department's responsibility to advise consumers on the quantity and variety of foods that will assure maximum benefit and satisfaction, research must continue on the nutritional requirements of persons of all age groups, on the nutrient and other values of foods, and on ways to conserve or enhance these values in household and institutional preparation and processing.

The kinds and amounts of foods consumed by different population groups and individuals must be determined periodically by surveys so that the nutritional adequacy of diets can be evaluated. Information on food consumption and dietary levels provides the guidelines needed for effective nutrition programs. This information also furnishes the basis for market analyses for different commodities and for development and evaluation of agricultural policies that relate to production, distribution, and consumer use of food.

USDA AND COOPERATIVE PROGRAM

The Department has a continuing program of research concerned with (1) nutritive and other consumer values of raw and processed foods as measured by chemical or physical means and by biologic response; (2) effects of household practices upon the nutritive values and inherent qualities of foods, and the development of improved procedures for household food preparation, care and preservation; (3) nutritional appraisal of food supplies and diets of different population groups; and (4) development of guidance materials for nutrition programs.

The research is carried out by two divisions of the Agricultural Research Service -- the Human Nutrition and the Consumer and Food Economics Research Divisions. Most of the work is done at Beltsville and Hyattsville, Maryland; some is done under cooperative, contract, or grant arrangements with State Experiment Stations, universities, medical schools, research institutes, and industry. The total Federal scientific effort devoted to research in these areas is 72.6 man-years. It is estimated that 11.1 scientific man-years are concerned with studies related to grain products.

Human metabolic studies and the related exploratory and confirmatory studies with experimental animals and microorganisms concerned with defining human requirements for nutrients and foods are not reported on a commodity basis, though some of the work is applicable to this report. This basic nutrition research represents a total Federal effort of 21.1 scientific man-years and is described in detail in the report of the Human Nutrition Research Division. Certain aspects of this research related to cereal products are considered briefly in this report.

PROGRESS -- USDA AND COOPERATIVE PROGRAMS

A. Wheat Products

1. Nutrient values. Research on the content of nutrients in a wide variety of wheats and wheat products continues at Beltsville and under research contracts with the American Institute of Baking at Chicago, Illinois, and with The Purdue Research Foundation at Lafayette, Indiana. Products are being studied from conventional and air classification flour milling practices and from conventional dough preparation and continuous rise dough procedures.

One hundred samples of 10 wheat products available to consumers, including white breads, whole wheat bread, rolls, flour, biscuit mix, doughnuts, and whole wheat cereals, were obtained in each of 10 cities (5 geographical areas) of the United States. Analyses are in progress for proximate composition, B-vitamins, amino acids, and some mineral elements (calcium, phosphorus, magnesium, iron, sodium, and potassium) at the American Institute of Baking. The samples were analyzed for 14 fatty acids in the extracted lipid fraction at Purdue University. At Beltsville, analyses are being made for reducing sugars, non-reducing sugars, starch, lactose, and pentosans. Existing methods were adapted for the analyses of wheat products and a manuscript giving the modifications and their analytical variability when used with wheat products has been accepted for publication in the Journal of the Association of Agricultural Chemists.

Procedures have been developed for separation of tocopherols and their analyses by gas chromatography. In whole wheat flour, epsilon tocopherol was the most prominent component; some alpha and beta tocopherols and to a lesser extent zeta-1 tocopherol were present. The quantitative extraction and purification of tocopherols of wheats and wheat products are currently being investigated. A paper on these studies was reported at the meeting of the American Oil Chemists Society in May 1966, and a manuscript is being prepared for publication.

In another phase of this study, samples of the hard wheat grain blends of 5 producing and milling areas, the bread flours from these grains, and the white breads made from the flours, by conventional and continuous dough mix, 20 samples in all, have been obtained. Twenty-eight of the 36 samples of soft and durum wheats, their flours, and products (white cake, crackers, or macaroni) also have been obtained.

2. Wheat fumigation. Research also was continued on possible nutritional implications of repeated use of fumigants on stored wheat. This study is being done in cooperation with the Market Quality Research Division. The wheat samples received treatment periodically during storage with methyl bromide, ethylene dichloride-carbon tetrachloride (3:1), and phosphine. Wheat has been in storage since the summer of 1965 and serious insect infestation in the control wheat has required changes in the fumigation schedule. Five periods, including the initial zero storage period, have been sampled to date. Some 475 samples of fumigated wheat, the untreated controls, their milling fractions (bran, shorts, low-grade flour, patent flour), the bread and roll doughs, and the baked breads and rolls have been received for analyses. To date, analyses for total solids, thiamine, riboflavin, and vitamin B6 components have not shown distinct differences attributable to effects of fumigation. Research was continued on the effects of fumigation of wheat during storage upon the baking performance of flour for household use and on eating quality characteristics of bread and rolls made from these flours.

3. Evaluation of wheat flours. To determine whether regional difference in flours was factor in satisfactory preparation of sauces and other dishes that depend on flour as a thickening agent, the viscosity of white sauces made with wheat flours from different geographical regions of the U. S. was studied. Two flours from the Southern region had greater thickening ability than most of the flours from the West, Southwest, and North Central Regions, as measured by viscometer readings of the sauces at 70° C. and at 24° C. Two agglomerated (instant) flours processed by the steam or hot water method made sauces that were often thinner than sauces made with flours of standard granulation. Sauces became more viscous with the use of fat or sodium chloride and fat in the formula, or with decreases in temperature.

The interchangeability of regular and agglomerated flours for consumer use was compared in different baked products. When the volume of agglomerated flour in the formula was reduced to approximate the same weight as regular flour, all but three baked products were equal in quality to those made with regular flour. Successful baked products were muffins, biscuits, waffles, coffee cake, plain cake, cream puffs, and cookies. Lower quality products were yeast rolls, popovers, and pastry made with agglomerated flour.

Physical tests are in progress on 26 flour samples representing all-purpose, bread, cake, cracker, and semolina flours from different sources of known history. Physical tests include amylograph, pH, mixograph, Hunter color, viscosity of a model system, starch swelling, and alpha-amylase.

4. Cracked wheat bulgur. Recommendations for preparation of cracked wheat bulgur were developed for use in consumer food programs such as School Lunch and Needy Family Programs. This food is new to many people in this country and requires special techniques in use to ensure palatable products because bulgur will become hard and tough or soft and soggy if prepared incorrectly. The amount of water used to cook bulgur was found to be basic in determining the kind of cooked product that resulted. Two parts of water to one part bulgur by volume made a cereal that was generally considered preferable to that made from three parts of water to one part of bulgur. Both flavor and texture of the product prepared with the smaller amount of water were preferred. Bulgur cooked in the oven or steamed was generally preferred to that cooked on top of the range.

B. Rice

As part of investigations of the nutritive value of convenience foods, proximate composition was obtained and energy values calculated for seven rice-containing foods in one or more market forms, both as purchased and ready-to-serve. Plain long-grain white rice (quick precooked, instant precooked, parboiled, and regular) as prepared to serve ranged in food energy per 100 gram portion from 94 to 139 calories. Regular rice was lowest in moisture (65.3 percent) and highest in protein (2.8 percent) among the market forms. Three brands each of canned and dry-pack Spanish rice and the product from a home recipe were compared. The ready-to-eat product from the dry-pack contributed higher energy value (average 106 calories per 100 grams) than the canned products (average 73 calories) or the Spanish rice from the home recipe (average 88 calories). The corresponding average protein values were 2.4, 1.3, and 1.8 percent. When prepared for serving, canned Spanish rice products were higher in moisture and lower in energy value, fat, and protein than were the home-prepared counterparts. Considerable variation in composition was noted among brands of a single product.

Medium-grain rice has been difficult for schools to use satisfactorily in their lunch programs because it became gummy when cooked by the usual procedures. Reducing the amount of cooking water $1/3$ cup per pound of Nova rice improved the texture of the rice cooked in the oven, a cooking method used extensively in quantity food production. This recommendation has been included in instructions prepared for the National School Lunch Program.

C. Tables of Food Composition

1. Vitamin E. A review of the vitamin E content of more than 5,000 food and feed items used for human and/or animal consumption was completed and published by the University of Wyoming at Laramie. This review was proposed and partially supported by the Human Nutrition Research Division. A total of 455 references were reviewed and fewer than 40 contained information on individual forms of tocopherols. The review was instigated by the increased recognition given to the importance of the tocopherols in metabolism of polyunsaturated fats and the extent of their use as naturally occurring antioxidants.

2. B-vitamins and trace elements. Summarization of data and derivation of representative values are nearing completion for a publication on the content of pantothenic acid, vitamin B₆ and vitamin B₁₂ in foods. The values will be given in terms of milligrams of the nutrient per 100 grams of edible portion and per 1 pound as purchased for each food item.

Also nearing completion is a preliminary table summarizing the data for 22 trace elements in foods arranged in 15 food groups. Data representing over 6,500 food samples analyzed for 1 to 22 trace elements have been reviewed, recorded on cards for punching and sorted by specific food. This table is particularly useful for indicating foods and food groups for which data are very limited or are conflicting.

D. Nutritional Evaluation of Cereal Products

1. Nutritional value of wheat protein. Work is continuing under contract on the nutritional value of wheat protein. At Michigan State University at East Lansing, the chemical analytical work has been completed in a study in which 12 healthy young men were maintained for 50 days on a controlled diet providing daily approximately 67 grams of protein of which over 90 percent was furnished by wheat products. The remainder of the protein was provided by fruit and vegetables. This research was reported at the Fourth International Cereal and Bread Congress, Vienna, May 22-27, 1966. Evaluation and interpretation of the data are continuing.

At the Agricultural and Technical College of North Carolina, Greensboro, data are being collected in contract research with young men on the nutritional value of wheat flour when fed alone or in combination with legumes or other cereals. The results obtained to date indicate that the young men were maintained in positive nitrogen balance on diets supplying 47 grams of plant protein per day when wheat alone or wheat supplemented by pinto beans, rice, or peanuts provided 75 percent (35 grams) of the dietary protein. No significant differences in nitrogen retention by the subjects were noted among the four diets. Serum urea levels fell, an observation which supports the data collected in Michigan. With the exception of lysine (which fell approximately 15 percent), plasma levels of essential amino acids did not change greatly with the wheat diets. The level of nonessential glutamic acid was more than doubled and levels of glycine and alanine rose appreciably. The decrease of alpha-amino-n-butyric acid to trace amounts in the plasma together with the detection of ethanolamine in the urine suggested to the researchers that with these wheat diets more methionine may have been used for synthesis of protein (rather than being metabolized) than was true with the self-selected meat-containing diets of the subjects. Portions of this research were reported in April 1966 at the Federation of American Societies for Experimental Biology in Atlantic City and at the American Association of Cereal Chemists in New York.

2. Nutritional value of carbohydrates. Work on the nutritional value of wheat starch as compared to sucrose is continuing at Beltsville. Ten young women, 19-25 years old, ate a controlled diet in which 85 percent of the carbohydrate was provided by wheat starch or by sucrose for 30-day periods. The most dramatic response to source of carbohydrate was noted in the levels of lactate dehydrogenase (LDH) in the blood serum. LDH is an enzyme involved in the conversion of carbohydrate to energy or body fat. Regardless of the sequence in which they ate the diets, 8 of the 10 women had elevated levels of this enzyme after eating the sugar diet for 30 days and depressed levels after eating the starch diet. Two subjects were unresponsive, indicating the possible functioning of genetic factors. Less dramatic but yet significant was the difference between serum levels of alkaline phosphatase (higher on the sugar diet than on the starch diet). An elevation in serum alkaline phosphatase is associated with bone tissue formation. Two other serum components which showed significant differences between diets were creatinine (higher on the sugar diet) and total fatty acids (lower on the sugar diet). No significant differences due to dietary carbohydrate were found in nitrogen and magnesium retention, fat excretion and digestibility, or in serum levels of glucose, urea, total protein or protein components, glutamic-oxalacetic and glutamic-pyruvic transaminase enzymes, cholesterol, phospholipids, and glycerides. Portions of these results were reported at the meetings of the Federation of American Societies for Experimental Biology in Atlantic City and of the American Association of Cereal Chemists in New York in April 1966.

A similar dietary study with 10 young men (19-23 years old) has been initiated. A number of additional measures of biological response will be included.

Scientists in Israel, supported by a PL 480 Grant from U.S.D.A., have reported evidence that the kind of carbohydrate may have a marked influence on the level of fat and cholesterol in the blood. Adult men and women of various ages, all prone to abnormally high levels of both triglycerides and cholesterol, were fed diets consisting of normal foods which supplied carbohydrates chiefly as sucrose or as starch. With the diets in which starch was the predominating carbohydrate, there was a precipitous drop in the blood triglycerides. Cholesterol levels followed a similar pattern but responded more slowly to dietary changes. Where sucrose predominated in the diet, marked elevation of blood triglycerides and cholesterol occurred.

3. Heredity and carbohydrate utilization. Research with human subjects and with experimental animals is rapidly providing evidence that the utilization of dietary carbohydrate may differ significantly with the type of carbohydrate. Research dealing with the response of two strains of rats, BHE and Wistar, also has indicated that heredity may be a factor contributing significantly to the response to various carbohydrates. For these strains of rats, comparison of the responses to age and fasting suggest that the metabolic activity of the liver may be a factor contributing to the shortened lifespan of the BHE rat fed sucrose.

The weight of the liver of the BHE rats fed sucrose increased with age, and was accompanied by a marked increase both in cholesterol and noncholesterol lipids. In contrast, the Wistar rats seemed to adapt with age to a high cholesterol diet. Liver weight remained unchanged and liver cholesterol decreased significantly. In spite of some relatively high liver lipids in 350 day old BHE rats compared with those found in Wistar rats, survival of BHE rats fed cornstarch or glucose (though not those fed sucrose) was similar to that of Wistar rats fed comparable diets. Liver cholesterol was high in the nonfasted BHE rat fed cornstarch but, in contrast to the sucrose fed rat, decreased on fasting along with a significant decrease in liver weight. In the fasted BHE rats fed cornstarch or glucose, neither cholesterol nor noncholesterol lipid changed with age. Two papers are being prepared for publication, one dealing with the influence of dietary carbohydrate on the composition of the liver and on some serum lipids of BHE and Wistar rats and one dealing with the results of microscopic examination of the tissue of these rats.

A project to be carried out under a recently negotiated research contract with the Hazleton Laboratories at Falls Church, Virginia, also is directed toward increasing our knowledge of the influence of heredity on the response to dietary carbohydrate. In this study fructose, in addition to sucrose and cornstarch, will be the carbohydrates fed. The effects of changing the type of carbohydrate at 150 days of age will also be investigated. An extensive series of blood measurements will be made in an attempt to find measurements in blood that will predict metabolic differences on some diets due to inherited characteristics.

4. Protein utilization on high rice diet. One way to extend scarce protein supplies is through improving physiological utilization. The relative economy of improving human nutrition with minimal increases in protein content of diets has been little explored. Under a PL 480 grant at Taiwan, Formosa, partial substitution of polished rice with sweetpotatoes has been studied in man and compared to supplementation with additional protein and with minerals and vitamins when fed to weanling rats.

Gain in weight of rats during 8 weeks was increased about 50 percent by sweetpotatoes replacing 10 percent of the rice calories, 100 percent by supplementation with standard mineral and vitamin mixtures, 200 percent by supplementation with soybean meal and fish flour (6 percent additional protein) and 250 percent by the protein and mineral-vitamin supplementation combined. Sweetpotatoes or the minerals-vitamins supplement each stimulated food intake. At first, feed efficiency (weight gain per unit of feed intake) was markedly higher on the protein supplement than on the sweetpotatoes or the minerals-vitamins supplement, but by the end of 8 weeks it was much lower and was similar on all diets except the basal unsupplemented rice.

Protein efficiency ratios (PER), (weight gain per gm of protein consumed) for 4 weeks and also for 8 weeks were higher with sweetpotato supplementation than without, regardless of whether the diet contained supplements of minerals and vitamins or of protein or of both. Protein efficiency ratios for the first 2 or 4 weeks exaggerated the benefits from protein supplementation alone, compared to 8 weeks PER levels from sweetpotatoes or from the minerals-vitamins supplement each without additional protein. The standard minerals-vitamins supplement did more to sustain steady high PER over the 8 weeks tested than did either the sweetpotatoes or the protein supplement to the basal diet which contained 93 percent rice by weight. These results on rat growth and others from the human metabolic studies indicate that protein supplements to human diets may be more or less wasted unless deficiencies in non-protein nutrients of the diets are also corrected.

5. Cholesterol synthesis and dietary fat. The body continuously synthesizes cholesterol, which is essential for normal function but which may be associated with certain pathological conditions. The rate of cholesterol synthesis may be significantly influenced by diet. In recent studies at Beltsville, the influence of type of fat on cholesterol synthesis was investigated. When rats were fed a diet containing 20 percent corn oil cholesterol synthesis was considerably greater than when the diet contained 20 percent beef tallow. In spite of marked differences in rate of synthesis, no differences were observed in the levels of cholesterol in the serum. A manuscript presenting these data has been submitted for publication.

6. Immunoproteins and protein status. A project has been initiated to determine the relationship of immunoproteins to protein status and amino acid intake. This project, to be carried out under a research grant with Iowa State University at Ames, will study antibody formation by rats fed proteins from various sources, such as corn, rice, wheat, soy, or egg. The proposed research should aid in establishing the sensitivity of antibody production to protein or selected amino acid deficiencies and further our understanding of the role of food proteins in maintaining a healthy and normally functioning body.

7. Nutrient interactions. Findings from studies to determine the nutritional value of various components of milk emphasize the importance of understanding possible interaction of various dietary components. The diets studied contained milk protein with various combinations of fat, as butter oil or corn oil, and carbohydrates, as lactose or cornstarch. When the diet included butter oil and lactose, calcium absorption by rats was much greater than with butter oil and cornstarch, corn oil and cornstarch, or corn oil and lactose. Retention of calcium, however, did not increase with the increased absorption; the excess calcium apparently was excreted in the urine. A high incidence of kidney stones occurred when the diets contained butter oil and lactose; no kidney stones were found with the diets containing starch. The incidence found with lactose and corn oil was low and did not differ significantly from the cornstarch diets. More research is needed to establish the mechanism responsible for the differences observed. A paper reporting the results of this research is being prepared for publication.

E. Food Consumption and Diet Appraisal

1. 1965 nationwide survey. Collection of data from the more than 15,000 households and 13,000 individual family members cooperating in the nationwide survey of food consumption in the United States is now complete. Tabulation of the data on households and preparation for tabulation of the data on individuals are in progress.

Preliminary review of the household data shows that family expenditures for food averaged \$33 a week in the spring of 1965. Of this, \$27 went for food bought and used at home, \$6 for meals and snacks eaten away from home. Home-produced and other foods for which no direct money outlay was made were valued at \$2. The money value of the food used averaged \$10.65 per person.

The total money value of food per family was 17 percent greater in the spring of 1965 than in 1955 when a similar survey was made. Most of the increase can be attributed to higher food prices but there was also a substantial increase in spending by farm families for food bought and eaten away from home. Their expenditures for eating out nearly doubled in the 10-year period and took 11 percent of total food money in 1965 compared to 7 percent in 1955. A report of the preliminary findings of the money value of food of households is being prepared.

Later, information will be available on the types and quantities of foods used by families in the spring of 1965. There will be information on approximately 250 foods--the percentages of families using the foods, average amounts and the money value of the food used. Where pertinent, data will be shown separately for purchased, home-produced, and Federally donated food. Publications will be prepared on dietary levels provided by the foods used. Similar information will be published also for the 4 seasons. For individual family members, data will be reported for about 20 different sex-age groups, for the U.S. total and probably for 2 regions.

2. Diets and nutriture of preschool children. A study was initiated to determine the nutritional situation of children, 2 and 3 years of age, in low-income families in Hawaii. Children from low-income families will be compared with those from higher income families with similar ethnic backgrounds. Biochemical, clinical, and psychomotor tests will be used to assess nutritional state. Correlations will be sought with social and economic characteristics of the child's family. The study will be done under a Cooperative Agreement by the University of Hawaii.

3. Household practices in homefreezer management. Preliminary review of the data collected during July 1964-April 1965 from 240 urban and 242 farm families in and near Fort Wayne, Indiana, showed that many homemakers had more than one reason for acquiring a homefreezer. About 40 percent of the reasons given by urban families were related to convenience--e.g., to have food on hand and to save shopping time. About 40 percent were related to economy--e.g., to buy meat by the side or quarter and to freeze local and

home grown fruits and vegetables in season. The reasons given by farm families were similar to those by urban families. Fewer urban than rural households (60 percent vs. 70 percent) kept the temperature of the storage areas in their freezers at the recommended temperature of 0° F. or below. Twenty-five percent of the freezers in urban households and about 35 percent of those in farm households were equipped with a thermometer.

Reports of these and other findings from the study are now being prepared as articles for publication in Family Economics Review. Preparation of a more comprehensive report will follow.

4. Nutritive value of national food supply. Estimates of food energy (calories) and selected nutrients provided by the per capita food supply are calculated each year from data on apparent civilian consumption, retail basis, developed by the Economic Research Service. The estimates indicate that for the past 10 years, the level of food energy has remained around 3,150 calories per capita per day--about 10 percent lower than in 1909-1913. This lower calorie level is the net result of about a 25 percent decrease in carbohydrate available for consumption, a 15 percent increase in available fat and a slight decrease in available protein, between 1909-1913 and 1965. This shift in the composition of the food supply caused the percentage of total calories furnished by carbohydrate to drop from 56 to 47 and the percentage of total calories furnished by fat to rise from 32 to 41. The percentage of total calories furnished by protein remained at about 12.

5. Nutritive content of school lunches. A nationwide study of the nutritive content of type A school lunches as served to sixth graders was initiated in cooperation with the School Lunch Division, Consumer and Marketing Service. Plans call for the collection and analysis of 20-lunch composites from 300 schools located in 19 states in five geographic regions. The objective is to provide data needed in evaluating the type A pattern. Because the pattern specifies the minimum amounts of foods required but does not specify maximum amounts that are allowed, the fat content of the lunches is of special interest.

F. Support for Food and Nutrition Programs

Developments in nutrition research continue to be studied and interpreted for application to problems in food selection and food use.

In anticipation of the expansion of child feeding programs, meal patterns for breakfasts and dinners suitable for children of all ages, were developed at the request of the School Lunch Division, Consumer and Marketing Service. These meal patterns will serve as guidelines for planning meals to meet the needs of children for food energy and the major nutrients. The patterns may be used independently or in conjunction with the type A lunch pattern.

To help promote better nutrition among low-income families, NCU food specialists and nutrition specialists cooperated with other Department agencies in the preparation of a teaching kit "Food for Thrifty Families." The kit consists of an adaptation of the "Daily Food Guide" and a series of 20 fliers that contain information on nutrition and simplified recipes for donated foods and food relatively low in cost. Cereal products included are cornmeal, corn grits, bulgur, rice, and rolled wheat.

The bimonthly publication of Nutrition Program News was continued. This publication reaches about 7,000 workers in nutrition and related fields. Talks to groups involved in community nutrition programs, radio tapes, and consultant help and participation in conferences contributed to coordinating and strengthening of nutrition programs.

Preliminary plans were made for the fifth National Nutrition Education Conference which will be held in February 1967. The conference is cosponsored by CFE and the Interagency Committee on Nutrition Education. The theme will be coordination and communication in nutrition programs.

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III. MARKETING AND ECONOMIC RESEARCH

GRAIN - MARKET QUALITY

Market Quality Research Division, ARS

Problem. Grain and cereal products are subject to damage, contamination, or deterioration in quality while in the marketing channels. These adverse effects may result from normal metabolic changes, the action of microorganisms, the attack of one or more of some 50 different kinds of common stored-product insects, or pesticide residues left by control measures applied against the insect pests. The damage may be conspicuously evident or insidiously hidden; may result in destruction of nutrient values or the presence of undesirable substances in food and feed; or insects may render products aesthetically unacceptable. Such deterioration affects the grade and price received, and end use, the wholesomeness of the product, and the competitive position of U. S. commodities in the world market. Research is needed to protect this multibillion dollar crop from outright destruction, evident deterioration, and hidden damage, especially as the need for food and animal feed becomes more critical throughout the world. The standard procedures that were formerly acceptable are becoming obsolete as a critical market demands higher quality, as international pesticide residue tolerances are in the process of being adopted, and as supply lines lengthen through overseas shipment in a worldwide distribution pattern. New methods of protection against insects and disease, of identifying and measuring quality changes, and means of maintaining quality over longer periods of time and under adverse conditions are some of the problems requiring attention.

USDA AND COOPERATIVE PROGRAM

The Department has a continuing program involving chemists, engineers and plant pathologists engaged in basic and applied research on the quality evaluation, quality maintenance and development of objective methods for quality evaluation of cereal grains. The research is conducted at Beltsville, Maryland and also by cooperative agreement with Purdue University, Lafayette, Indiana and by a research grant with the University of Minnesota.

The Federal scientific effort in this area totals 16.7 scientific man-years: quality evaluation, 14.7, and quality maintenance 2.0.

PL 480 projects include a grant to the Institute for Cereals, Flour and Bread, T. N. O., Wageningen, Holland, providing for the development of simple, reliable and accurate tests to determine relative hardness of wheat for the purpose of devising a system of classifying wheat for use in official grading and inspection. Its duration is four years (1965-1969) and involves \$70,670 in Dutch guilders.

A grant to the Instituto Nacional de Investigaciones Agronomicas, Madrid, Spain, provides for a study to develop and evaluate equipment and methods for determining the proportions of durum (semolina) and common wheat (farina) in macaroni and spaghetti products. Its duration is five years (1964-1969) and involves \$43,563 in Spanish pesetas.

There is also a continuing program involving entomologists and chemists engaged in basic and applied research on problems of insect infestation, damage, and contamination of grains and cereal products in the marketing channels. The research is conducted at Manhattan, Kansas, and Tifton, Georgia, in cooperation with the respective State Agricultural Experiment Stations, the Transportation and Facilities Research Division, the Agricultural Stabilization and Conservation Service, farmers' cooperatives, the Association of Operative Millers, the American Corn Millers Federation and other industry groups. The Commodity Credit Corporation makes available various commodities and storage facilities for experimental use, thus greatly facilitating the program. There is also overall cooperation with several State Agricultural Experiment Stations through participation in Regional Project WM-52, "Maintaining marketability of stored grain and cereal products through insect control by methods leaving no, or a minimum of, pesticide residues."

A three-year cooperative agreement with the Kansas State University became effective in December 1965, for research on the biology and behavior of the Angoumois grain moth.

A grant to the Administration of Agricultural Reserves and Surpluses, Montevideo, Uruguay for a five-year study of underground and aboveground storage of corn in airtight silos in relation to maintaining quality and preventing insect infestation during long-term storage. It continues until May 1967 and involves PL 480 funds with a \$73,042 equivalent in Uruguayan pesos.

A grant to the Indian Agricultural Research Institute, New Delhi, India, for a three-year study of varietal resistance of wheat kernels to damage by the rice weevil and lesser grain borer. It continues until February 1968 and involves PL 480 funds with a \$15,146 equivalent in Indian rupees.

A grant to the Tokyo University of Agriculture, Tokyo, Japan for a three-year study, part of which is on the constituents of wheat that attract insects. It continues until August 1968 and involves PL 480 funds with a \$38,622 equivalent in Japanese yen.

A grant to the Hebrew University in Jerusalem, Israel, for a five-year study on the biology of the khapra beetle. It continues until October 1966 and involves PL 480 funds with a \$97,123 equivalent in Israeli pounds.

A grant to the Maharaja Sayajirao University, Baroda, India, for a five-year basic research study on the physiology of fat metabolism in relation to diapause in the khapra beetle. It continues until January 1970 and involves PL 480 funds with a \$33,907 equivalent in Indian rupees.

A grant to the University of Helsinki in Finland, for a five-year study on the effects of pesticides on plant commodities. Part of this will deal with malathion in wheat. The project continues until December 1969 and involves PL 480 funds with a \$96,441 equivalent in new Finnish finnmaks.

A grant to the Hebrew University in Rehovot, Israel, for a five-year study on the effect of ethylene dibromide fumigant on farm animals. It continues through August 1966 and involves PL 480 funds with a \$117,442 equivalent in Israeli pounds.

The Federal effort devoted to research on the prevention of insect infestation totals 7.7 scientific man-years, of which 0.1 was under a cooperative agreement.

Line Project MQ 1-31, a study on preconditioning of insects to fumigation, was terminated because the scientist resigned and no qualified replacement was available.

Line Project MQ 1-18, a study of the effects of air movement on stored-grain insects, was discontinued in July 1965.

PL 480 project E15-AMS-9(a), a study in Italy on insect infestation in pastas, was terminated upon successful completion of the plan of work.

PROGRAMS OF STATE EXPERIMENT STATIONS

A total of 18 scientist man-years is devoted to this area of research.

REPORT OF PROGRESS FOR USDA AND COOPERATIVE PROGRAMS

A. Objective Measurement and Evaluation of Quality

1. Quality Indicators for Stored Wheat. This study on objective measurements to indicate quality changes in stored wheat was completed. The study comprised four hard red spring and four hard red winter wheat varieties adjusted to 12, 14, and 16 percent moisture content and stored for various periods at temperatures of 50, 75, or 86°F. The wheats stored at the highest temperature and highest moisture content generally changed in quality at the fastest rate. The data has been tabulated and statistical analysis to determine the significance of each of the quality measurements and their inter-relationships is underway.

2. Sampling Research. A series of tests on the performance of three commercially available grain samplers and a manually operated pelican sampler was completed. About 5,000 samples were drawn by the devices and a separate analysis performed on each. The analysis data are awaiting statistical study.

3. Flour-Yielding Capacity of Wheat. Measurements related to the bran content of wheat have been found to be significant as indicators to flour yield in four classes of wheat. Computer analysis of our earlier data has pointed to the three most important indicators of flour yield in each class of wheat. In hard red winter: test weight, pentosan content, and mill bran account for 92 percent of the variability of flour yield. In hard red spring: test weight, pearling index, and mill bran account for 77 percent. In soft red winter: pearling index, kernel density and mill bran account for 93 percent. In white wheat: pearling index, pentosan content and mill bran account for 86 percent. Procedures combining two or more of these factors to predict flour yield will be investigated.

Rapid milling procedures, using the Brabender Quadrumat, Jr. mill, also predicted flour yield of hard red winter wheat but in the soft red winter, hard red spring, and white club wheats, the Quadrumat mill bran yield appeared to be a better indicator.

4. Protein Content of Feed Grains. A rapid technique was developed for determining protein content in grain sorghums by measuring the change in reflectance upon adding water to the sample. The reflectance measurement was made at 640 m μ on 350 gram samples (total 157 samples) of bulk grain sorghums free from foreign material contained in a 30 square-inch sample viewing cell. This large sample viewing area permits measurement of grain sorghums which are non-homogeneous and parti-colored products with a capability of detecting 0.05 percent difference in reflectance between samples.

The dye binding test for measuring protein content in wheat was adapted to grain sorghums, barley, corn, soybean meal, cottonseed and alfalfa meal. Conversion tables with moisture corrections were prepared for each commodity.

5. Detection of Mixed Corn. Hysteresis effects make it possible to detect lots of corn resulting from the mixing of high moisture corn with artificially dried corn. The range of moisture contents of individual kernels is found to be greater in mixtures. The greater the drying temperature, the greater is the range of moisture contents. An electronic instrument is now being built which will make rapid determinations of the moisture contents of individual kernels. Heat drying of corn causes changes in the solubility of the proteins in certain solvents. Extraction of the protein from the ground corn and determination of the protein in the extract by a colorimetric method offers possibilities as a means of detecting mixtures. This procedure is being investigated.

6. Aflatoxin in Corn. Aflatoxin in corn is apparently associated with heat-damaged kernels. Aflatoxin was found in about 60 percent of individual heat-damaged kernels tested, but was not found in other types of damage ordinarily found in kernels of corn, such as mold, cob rot, weathered, ground, silk cut, sprouting and insects. Because of the characteristic dark red color of heat damage, the individual kernels were identified by using a simple ratio absorbance measurement at 475 and 575m μ . This absorbance technique for identifying heat damage in kernels of corn agreed with the Grain Inspector's "eye-ball" test in all 67 kernels tested.

7. Quality Evaluation of All-Purpose Flour. The Perten falling number test showed considerable promise in evaluating the quality of all-purpose flour. Plans are being made by the Grain Division, Consumer and Marketing Service, (a collaborator in this project) to conduct a collaborative study of the Perten (a viscosity test) falling number determination among a selected group of cereal chemists in the United States. Large samples of all-purpose flour produced for consumer use by mills providing all-purpose flour to the Government are being obtained for additional chemical, physical and baking (bread, biscuits, cookies, etc.) tests.

8. Macaroni and Spaghetti Products. A new chromatographic-colorimetric method, a modification of the Matweef method for measuring the different proportions of common and durum wheat flours in finished macaroni products by the presence of the sitosteryl palmitate, has shown promising results. The new method is satisfactory for detecting even low amounts of this chemical component in macaroni products. Samples of U. S. grown common (hard red winter) and durum wheats have been sent to the investigators to supplement the Spanish wheats used initially in this study.

B. Quality Maintenance in Storage

1. Mycotoxin Fungi in Stored Corn. Two studies relating to the development of toxin-producing molds are currently underway. They consist of: (1) storage of high moisture field shelled corn (18 percent moisture) aerated to maintain temperatures at ambient, 30, 40, and 50°F.; (2) corn dried by the drier process and handled in various ways in an attempt to eliminate the water condensation on the peripheral portions of the grain, associated with this method of drying.

After five months storage, there was little mold growth and no Aspergillus flavus in the corn stored at initial moisture contents of 18 percent. An exception was at the surface of the grain stored at 50°F. where the seed moisture had increased to 20 percent and was invaded by Penicillium spp.

Corn subjected to drier, if not removed from the holding (tempering) bin even if aerated at low rates, 0.1 - 0.25 CFM/bu, molded heavily at the surface of the grain and next to the bin wall where moisture had accumulated.

Species of Penicillium were most common in all four bins examined with Aspergillus flavus and Aspergillus fumigatus occurring in one bin. Samples of dryerated corn transferred from holding bins were not molded.

2. Quality Changes Due to Insecticides. The effect of Cab-o-sil, Silica Aerogel 68, Perma Guard, Kenite, and Malathion on test weight per bushel and yield of flour of hard winter wheat after two years' storage is unchanged from results previously reported on one-year storage. For all treatments, except Malathion, there is an initial but no subsequent decrease in test weight. Flour yield is unaffected. The Cab-o-sil, Silica Aerogel 68 and Perma Guard treated samples of hard winter wheat increased in fat acidity values to about the same level as the control. Increases were less for the Kenite and Malathion treated samples. Marked decreases were shown in the mixing tolerance values by the farinograph for the control and four of the insecticide treated samples, but this dough property increased for the Cab-o-sil treated wheat. The breads from the Perma Guard, Kenite and Malathion treated samples decreased in loaf volume (rich formula method without dry skin milk) during the two-year storage period.

In a cooperative study with the Stored-Products Insects Branch, Market Quality Research Division and the Human Nutrition Research Division on quality changes due to fumigation, two controls (identified as outside and cold) and three fumigated wheat samples treated with (1) phosphine; (2) methyl bromide, and (3) three parts of ethylene dichloride plus one part carbon tetrachloride comprise the samples in this study. No changes have taken place in the chemical and milling characteristics at the end of nine months storage as compared with the initial tests. The absorption and mixing peak values, according to the farinograph tests, increased gradually with storage time for the fumigated and control samples. Fumigation treatment improved the dough mixing tolerance, another important quality property, to a greater degree than the control samples. Phosphine and methyl bromide treated samples were best of the group in this respect after nine months storage. There were no significant differences in bread loaf volume between the controls or the fumigated wheats. Bread loaf volume, crumb grain and color decreased with length of storage time; the losses were small and gradual and about of equal magnitude for the fumigated as the control samples. The hot breads from the fumigated samples were normal in odor.

3. Microflora of Wheat. The final report on this project presents considerable data on the biochemical and technological properties of stored soft wheat and flour milled therefrom. Physical properties of the doughs are stated to improve with storage resulting in better loaf volumes. Diastatic activity (maltose) of the grain underwent a gradual decrease under all conditions of storage. A positive correlation between fat acidity values and loaf volume was found in 8 of 12 cases involving wheat stored under different conditions of temperature and relative humidities. Stability of the flour was found to depend not only upon the conditions of storage

but also upon the mode of preparatory treatment of the grain prior to milling. Leveling off and/or a decrease in fat acidity values with time of storage in some of the Polish studies is explained as related to development of molds.

C. Prevention of Insect Infestation

1. Biological and Physical Control. In tests with corn in 500-bushel bins in Georgia a silica aerogel dust continued to give protection comparable with that from malathion 32 months after treatment. The effectiveness of both materials decreased considerably between the 24- and 32-month inspections. Another silica aerogel and two diatomaceous earths were less effective. The lesser grain borer was the predominant insect in the inert dust-treated corn and the rice weevil in the malathion treatment. Malathion residues were 1.2 ppm at 24 months and 0.9 ppm at 32 months, indicating the treatment has reached the breaking point of protection. Bioassay tests showed that the inert dusts were only about half as repellent at 24 months as at 12 months. In field tests in Kansas, where thirty 3,250 bushel-bins of wheat were sampled regularly over a 30-month period, observations indicated that diatomaceous earth and silica aerogel wheat treatments were nearly as effective as malathion in preventing insect buildup in dry wheat. Observations also indicated that the inert dust treated wheat tended to retain moisture from leaks during snow and rain storms, causing spoilage and insect development in localized areas.

Adherence of inert dust to grains during treatment in the laboratory depends on kind of dust and variety of grain. Grain sorghum and wheat retain equal amounts of the same dust while less remains on shelled corn. Silica aerogels and diatomaceous earths were evaluated. Toxicity studies with 12 percent moisture wheat, which was aged for 9 months following treatment with 2 diatomaceous earths, Perma Guard and Kenite 2-I, showed kills of rice weevils comparable to newly-treated wheat. The repellent action of these two materials on wheat was not measurably reduced after 24 months of aging.

Ninety-seven selected double crosses of corn with F44 as one of the parents, were rated for rice weevil resistance during storage. Lines crossed with F44 as the female parent were considerably more resistant to rice weevil invasion than were reciprocal crosses of the same lines. When F44 was used as the female parent, no rice weevil progeny developed on nine of the 65 crosses. The number of progeny from the others averaged 0.2 to 8.2. Average progeny from reciprocal crosses averaged 1.6 to 52.6. In tests with 65 crosses with F6 as one parent the resistance was not as great, no progeny from only one cross and ranging to a high of 28.0. Average progeny from crosses of some of these lines ranged from 1.6 to 75.8 rice weevils. In 70 crosses with GT 112 as one parent there was even less resistance, progeny averaging 4.6 to 99.0. In tests with 49 varieties grown in Corn Performance Field Crops Variety Trials, none of the hybrids was resistant to rice weevils, progeny averaging 87.0 to 282.0. (Cooperation with Georgia Agricultural Experiment Station)

The final report from the study of insect infestations in spaghetti, macaroni, noodles and other pastas lists seven significant findings or accomplishments. These are: (1) the process of milling wheat destroys internal infestations; (2) Sitophilus oryzae and S. granarius cannot subsist in flour; (3) the pressure to which dough is subjected in modern pasta presses is sufficient to kill Sitophilus sp. eggs; (4) the high temperatures inside pasta driers are sufficient to kill eggs that are present and to prevent egg-laying; (5) infestation can occur only after the pasta is made; (6) pasta may become infested while stored at the mill, during warehouse storage, in transit, and while on the grocery shelf, and (7) valuable contributions were made to insect-rearing methods and on the biology of the two species of Sitophilus. The project has been satisfactorily completed and three articles published reporting the results.

2. Biology, Ecology, and Physiology. Nine different antibiotics tested individually in several percentages in the diet of khapra beetle larvae had varying effects on length of larval development, from none to a considerable delay in development with the lower percentages of tetracyclin and tyrothricin. A pheromone was collected from virgin female adult khapra beetles by extraction in diethyl ether or by allowing the females to crawl on filter papers. Both sexes of the beetle are attracted by the pheromone, classing it as an aggregating substance rather than a sex attractant. Virgin females were more strongly attracted than were virgin males or females after copulation. Preliminary tests showed that larvae of the khapra beetle detect light in the range from red to violet. Extensive tests have been conducted and much has been learned about response to different wave lengths and intensities of light and this work is continuing. Khapra beetle adults preferred the dry end of a long humidity gradient chamber. From 54 to 60 percent gathered in the end with a range from 0 to 10 percent relative humidity. The response varied with sex and age. In temperature gradient studies, 70 to 80 percent of the adults preferred the range between 78 and 88°F., although about 15 percent were in the area of 96 to 98°F.

A temperature of 95°F., a relative humidity of 70 percent, and crushed wheat as a rearing medium were found to be the most suitable conditions for obtaining non-diapausing specimens of the khapra beetle in a fat-metabolism study. The fat body of 5th-instar larvae contains large amounts of lipids and some albuminoid-like globules. No uric acid was demonstrated in the fat body at this stage. During the course of this study, a protozoan was found to parasitize the khapra beetle larvae, causing considerable mortality. This organism might well be a potential agent for control of the insect.

3. Improved Pesticidal Control. Chloropicrin fumigation by the recirculation method of application was highly effective in a test conducted in a large flat storage building containing 100,000 bushels of wheat. The dosage applied was 2 pounds/1,000 cubic feet, recirculated for two hours. Average mortality of test insects in 112 locations was 94.8 percent for adult confused flour beetles and 99.2 percent for immature rice weevils. The

principal areas of insect survival were midway between the aeration ducts at each end of the building. Static pressure readings showed there was little air movement in those locations. Gas chromatographic analyses of air samples from the same locations revealed low chloropicrin concentrations there, but good distribution elsewhere. There was no evidence of the gas settling out of the grain during 24 hours after application.

Newly combined wheat with a moisture content of 13 to 14 percent was effectively protected against insect infestation by malathion or diatomaceous earth during the first 12 months of storage in 100 bushel wooden farm-type storage bins in Kansas.

The effectiveness and persistence of a fresh malathion treatment on wheat was not altered by aeration for five weeks at conventional airflow rates of 0.05 or 0.1 c.f.m./bushel, with the air at 80°F. and 75 percent relative humidity. When the wheat had been treated with malathion four months prior to aeration, effectiveness of the malation declined after only two weeks of aeration. Air drawn from malathion-treated wheat was highly toxic to rice weevils at first and progressively declined in toxicity during five weeks of aeration at conventional airflow rates. Air drawn at a high flow rate remained nearly as toxic to rice weevils after four weeks of aeration as it was during the first week.

In preliminary tests, two treatments show promise as economical and effective means of dealing with the difficult problem of insect infestation in rail cars. For empty cars, one quart of a mixture containing 2½ percent of malathion and 2½ percent of dichlorvos is applied with 100 p.s.i. of air pressure to make a heavy, wet fog. It is directed behind the car liners and onto the inner surfaces of the car. The pressure blasts the spray into almost impenetrable cracks and crevices. Total time for treatment and aeration is 20 minutes per car and the material costs less than 60 cents. So far, 100 percent of test insects have been killed. For rail cars loaded with packaged cereal products and for loaded bulk hopper car, fumigation with aluminum phosphide pellets at the rate of 165 per 1,000 cubic foot has given excellent results.

Two tests conducted in quonset huts each containing 18,000 bushels of shelled yellow corn showed that fumigation can be accomplished by putting aluminum phosphide tablets on the surface of the grain rather than probing them in by the conventional method. The dosage rate was 90 tablets per 1,000 bushels and in one test there was 93.8 percent mortality of immature rice weevils and 99.9 percent mortality of adult red flour beetles.

Aluminum phosphide pellets at 165 per 1,000 cubic foot and tablets at 45 per 1,000 cubic foot have excellent control of insects inside packaged food products stacked on pallets and covered with polyethylene sheets for fumigation. Several common types of packages were included, some of which were plastic films and foil-laminated over wraps. There was 100 percent

mortality of rice weevil adults, confused flour and saw-toothed grain beetle adults, dermestid larvae, and Indian-meal moth larvae. The kill of immature rice weevils was only about 99.9 percent but this stage would not infest most packaged food products.

By utilizing air movement, malathion aerosol in quantities lethal to insects has been pulled down through as much as 14 feet of shelled corn. Compaction of the grain influences airflow and rate of airflow in turn affects the distribution of the aerosol through the grain mass.

In a study to determine the effect of ethylene-dibromide-(EDB)-fumigated feed on animals, analytical methods developed on the projects using minute quantities of sample were used to establish curves of aeration time required for a fumigant mixture applied to wheat, barley, and corn. It appears that the effect of EDB on laying hens was not related to gonadotropin deficiency since treating hens with a follicle-stimulating hormone did not improve depressed egg weights. The chemical composition of small eggs from hens fed on EDB-fumigated feed was similar to normal eggs as far as total nitrogen, calcium, phosphorus and fat were concerned. Only the total amounts of yolk constituents seem to be affected. The semen apparently is not affected by treatment of cocks with EDB. The influence on bull semen was stronger when the EDB was administered in double doses every second day, than in single daily doses. In the study of acute poisoning of rats by EDB, the structure of the main metabolites in urine was clarified.

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Objective Measurement and Evaluation of Quality

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RICE - MARKET QUALITY

Market Quality Research Division, ARS

Problem. Harvested rice is subject to damage or deterioration in quality while in marketing channels through normal metabolic changes, by the action of disease organisms, and by insect infestation. There is need for developing more effective ways of preventing insect infestation during storage, handling, processing, packaging, and transportation of rice. Attention must be given to finding control methods that will minimize or eliminate pesticide residue hazards. To maintain the quality of rice, more precise information is needed on the changes that occur in handling, storage, and transportation. To insure uniform and standardized products and more equitable prices to all concerned, new and improved procedures for measuring quality factors must be developed for use in inspection, grading, and standardization operations.

USDA AND COOPERATIVE PROGRAM

The Department has a continuing program involving engineers, chemists, and plant pathologists in basic and applied research on the quality evaluation and quality maintenance of rice. This work is located at College Station, Texas, in cooperation with the Texas Agricultural Experiment Station.

The Federal scientific effort in this area totals 2.0 scientific man-years: quality evaluation 1.0, and quality maintenance 1.0.

A grant to the Department of Plant Chemistry, Valencia, Spain, provides for a study on storage changes in milled rice and their relation to market quality. Its duration is for 4 years, 1964-1968, and involves P.L. 480 funds with a \$62,479 equivalent in Spanish pesetas.

A grant to the National Institute of Hygienic Sciences, Tokyo, Japan, provides for a study covering the cause and development of mycotoxins in rice as a result of invasion and growth of fungi during postharvest conditioning, handling, and storage. Its duration is 3 years (1965-1968) and involves \$33,164 in Japanese yen.

There is also a continuing program of basic and applied entomological research directed toward the prevention of insect infestation in rice in the marketing channels, headquartered in Fresno, California. The Federal effort was temporarily reduced to only 0.5 scientist man-year by assignment of personnel to research on gamma irradiation, which is also pertinent to insect infestation in rice. Much of the cross-commodity research at Savannah, Georgia, reported in Area 13, "Insect Control in Marketing Channels," is applicable to the problems in rice.

A grant was made to the Tokyo University of Agriculture, Tokyo, Japan, for a 3-year study, part of which is on the constituents of rice that attract insects. It became effective in August 1965 and involves P.L. 480 funds with a \$38,622 equivalent in Japanese yen.

PROGRAM OF STATE EXPERIMENT STATIONS

A total of 2 scientist man-years is devoted to this area of research.

REPORT OF PROGRESS FOR USDA AND COOPERATIVE PROGRAMS

A. Objective measurement and evaluation of quality

1. Degree of Milling and Color of Rice. A commercial transmittance instrument, the "Ratiospect," was modified by adding a reflectance attachment and special optical unit. The optical unit is arranged to permit either reflectance or transmittance measurements of a rice sample to be made by moving a selector lever. Degree of milling, degree of parboiling and color measurements made on a large number of graded rice samples with the modified "Ratiospect" show that correlation between instrument readings and visual grades were high for white milled rice but somewhat lower for parboiled rice because of interactions between degree of milling and degree of parboiling.

2. Environmental Conditions and Stress-Cracks in Milled Rice.

Instrumentation and techniques were developed to measure stress-crack damage in milled rice caused by environmental changes (temperature and relative humidity). The results showed the following: (1) high-moisture rice is more susceptible to damage than low-moisture rice; (2) rice subjected to a relatively small change in relative humidity (20 percent) is severely damaged when the temperature of the new atmosphere is relatively high, 30° C. or more, and (3) the amount and rate of damage is directly related to the magnitude of the change in moisture content the rice would undergo if allowed to come into equilibrium. Varieties differ greatly in their susceptibility to stress-crack damage, from a relative value of one for Century Patna 231 to a value of over 12 for Blue Belle.

B. Quality maintenance in conditioning, handling and storage

1. Microbiological, chemical, and Physical Deterioration of Rough Rice.

The relation between storage relative humidity and the rate of establishment of infection was studied by inoculation of rough rice with the dry spores of an isolate of the Aspergillus flavus group and subsequent storage at 30° C. and relative humidities ranging from 70 to 90 percent. Infection occurred at a high rate at all relative humidities during the first week of storage. Competition by other fungi had little or no effect on the prevalence of infection by A. flavus in the rice stored in relative humidities of 70, 73, and 75 percent for periods up to 4 weeks. Members of the A. glaucus group

became increasingly competitive as the storage relative humidity was increased from 80 to 90 percent. A. candidus became increasingly competitive after 21 days of storage at 87 percent R. H. and after 14 days at 90 percent. The prevalence of Penicillium spp. also started to increase rapidly after 21 days of storage at 90 percent.

These data provide additional evidence that prevalence of fungi in rough rice as determined by isolations from kernels or colony counts by the dilution method is an inadequate method of estimating the effect of fungal invasions on deterioration or changes in quality. Studies to relate the rate of CO₂ evolution with fungal induced changes in quality during storage are in progress.

2. Heat Damage of Rice. Asperigillus fumigatus was investigated as a possible cause of heat damage. Although the fungus grew well on rough rice at temperatures ranging from 40 to 50° C. and caused a general deterioration of the grain, heat damage symptoms or other distinctive kernel discoloration did not develop. Rough rice inoculated with A. parasiticus developed some heat damage symptoms but all tests gave widely variable results and must be considered as inconclusive.

3. Damage and Off-Color in Rough Rice. Discolored bran in second-cutting Belle Patna rice was found to be caused by infection by Helminthosporium oryzae. The somewhat atypical discoloration appeared to result from infections initiated in kernels in the latter stage of maturity.

4. Storage Changes in Milled Rice. A highly significant positive correlation has been found between the disulphide groups content and the quality of milled rice. This relationship is evident in both fresh and stored rice. Microbiological respiratory activity was not indicated as a major cause of changes in quality in milled rice during storage. Preliminary data show that gelatinization and pasting characteristics are not useful for evaluating cooking quality changes in rice during storage. The N index of objective measurement of quality is a valid method of evaluating stored as well as fresh milled rice.

5. Mycotoxins in Rice. The production of aflatoxins in high moisture rough rice was studied in early, mid-season, and late harvested rice in cooperation with the T&F Division. These tests clearly showed that all other factors being equal, aflatoxins can be expected to develop most rapidly in high moisture rice held under the higher temperatures prevailing during the early harvest. Extensive investigations in the laboratory of the effect of temperature in the development of aflatoxins in rough rice have supplied verification. Although there are some slight differences in response to temperature among various strains of the A. flavus group, optimum temperature for aflatoxin production ranges from 20 to 35° C. Moreover, the most important aspect of the temperature/aflatoxin relationship is that production and accumulation increases sharply as the temperature increases within the optimum range. As an example, 6 days at 20° C. were required for aflatoxin concentrations to reach the level obtained in 2 days at 30° C. in one such test.

About 170 isolates of fungi have been obtained from 136 samples of milled and rough rice from 92 locations in Japan. Preliminary tests of acute toxicity of culture extracts have been made by oral administration of .02 ml of an extract to two-week-old mice. Some strains of Aspergillus flavus, A. ochraceus, A. clavatus, and Penicillium islandicum were characterized by high toxicity in this test.

C. Prevention of insect infestation

1. Biological and Physical Control. Increasing the intensity of infrared radiation applied to rice internally infested with immature stages of the rice weevil, lesser grain borer, and Angoumois grain moth caused a proportionate increase in insect mortality, even though the total dosage of radiation was held at the same level.

Rice weevils and lesser grain borers showed a definite preference for feeding on certain varieties of rice in tests where the results were based on an evaluation of the amount of damage to kernels.

In olfactometer tests rice weevils were more strongly attracted to polished rice than to rough rice. The attracting substances were extracted more readily with methanol than with ether. The methanol extracts from rice bran were more strongly active than the ether extracts.

PUBLICATIONS REPORTING RESULTS OF USDA AND COOPERATIVE RESEARCH

Objective Measurement and Evaluation of Quality

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Quality Maintenance in Conditioning, Handling and Storage

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PUBLICATIONS -- STATE EXPERIMENT STATIONS
and COOPERATIVE PROGRAMS

A. Objective Measurement and Evaluation of Quality

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FEED AND SEED - MARKET QUALITY
Market Quality Research Division, ARS

Problem. Many methods of determining seed and feed quality currently in use require too many man-hours, impose tedious work on the analyst, are incapable of high degrees of standardization, and do not provide accurate indices of quality. Practical methods are needed for determination of such quality factors as mechanical purity, genetic purity, germination, vigor, weed seed content, protein content (of feeds) and infection with disease organisms. The deleterious effects of high temperatures and relative humidities on stored seed are well known but little is known about the part played by storage molds, especially the minimum temperature-relative humidity combinations under which the storage molds survive in stored seed. There is urgent need to increase basic research which would serve as a basis for developing more practical methods of determining seed and feed quality and for recommending improved practices of storing seed.

USDA AND COOPERATIVE PROGRAM

The Department has a continuing long-time program on seed research involving botanists, plant physiologists, plant pathologists, engineers, and chemists engaged in both basic and applied research on quality evaluation and quality maintenance of seed. This research is conducted at Beltsville, Maryland, and College Station, Texas, and by research contract with the Oregon Agricultural Experiment Station and by cooperative agreement with Mississippi State University.

The Federal effort in this area totals 6.2 scientific man-years: quality evaluation, 5.5, and quality maintenance, 0.7.

A PL 480 grant with the Instituto Biologico, Sao Paulo, Brazil, provides for a study of substrate moisture levels for germination testing of agricultural seeds. The project runs from 1961 to 1966 and involves \$31,016 equivalent in Brazilian cruzeros.

A PL 480 grant with Rijksproef-station, Wageningen, Netherlands, provides for a study of the health condition of seeds in commercial channels and development of methods suitable for routine testing for seedborne organisms. The duration of the project is five years, beginning 1963, and the total grant in Dutch guilders is the equivalent of \$55,918.

A PL 480 grant with Samenprüfstelle, Munster, Germany, provides for a study of the biological and environmental factors affecting the physiological maturity of grass seeds. The duration of the project is three years, beginning April 1965, and the total grant in German marks is the equivalent of \$31,775.

A PL 480 grant with Forschungsgemeinschaft für Saatgutforschung, Reutlingen, Germany, provides for a study of methods for maintaining the germination of seeds in storage and in trade channels. The project has a duration of five years, beginning April 1965, and the grant in German marks is the equivalent of \$52,338.

A PL 480 grant with the Agricultural Research Station, Beit Dagen, Israel, provides for research to find a satisfactory invisible marker of seeds in commercial channels and for research purposes. The project runs for three years, beginning February 1965, and the total grant in Israeli pounds is the equivalent of \$45,640.

A PL 480 grant with the Indian Agricultural Research Institute, New Delhi, India, provides for a study to evaluate the X-ray technique for detecting empty seeds in purity testing and for determining seed viability. Its duration is five years, beginning October 1965, and the total grant in Indian rupees is the equivalent of \$37,464.

A PL 480 grant with the Weizmann Institute of Science, Rehovoth, Israel, and the Hebrew University, Jerusalem, Israel, provides for research to isolate and determine the structure of germination inhibitors in seeds. The project runs for three years, beginning October 1964, and the total grant in Israeli pounds is the equivalent of \$50,722.

A PL 480 grant with the Israel Institute of Technology, Haifa, Israel, provided for a study to develop tests for nutritive value of cereals grains and feeds. The duration of the original grant was four years, 1961-1965, but an extension of one additional year was approved. The total grant involved an expenditure of \$53,298 equivalent in Israeli pounds.

PROGRAM OF STATE EXPERIMENT STATIONS

A total of 4 scientist man-years is devoted to this area of research.

REPORT OF PROGRESS FOR USDA AND COOPERATIVE PROGRAMS

A. Objective Measurement and Evaluation of Quality

1. Development and Standardization of Seed Testing Equipment. Laboratory models of a seed blower, for semi-automation use, utilizing the Venturi principle, and a seed subsampling device with an air-operated seed transfer sleeve for recycling all seed not drawn off in the subsample were designed

and constructed. Of several air measuring devices used to calibrate a seed blower for precision separations, a thermo-anemometer was the only one with potential value but it was not completely successful.

2. Seed Metabolism. By kinetic analysis in vivo and in vitro, it was shown that the "activation" of ribosomes parallels for formation of polysomes throughout the early phases of wheat embryo germination. The initial reaction is extremely rapid and manifests itself approximately ten minutes after water uptake. This "trigger" reaction has been obtained in vitro and may be described as an ATP-requiring messenger RNA-ribosome binding system.

3. Determining the Purity of Certain Grass Seeds. Methods of overcoming the detrimental effect on germination of some grass seeds caused by solvents used in the flotation purity analysis were studied by the following methods: (1) Froth flotation using a special flotation cell; (2) Use of supplemental drying treatments, and (3) Use of low-surface tension aqueous solution for flotation. Promising results were obtained with a combination of these methods, but further tests are needed for conclusive results.

4. Hay Moisture Meter. Good results were obtained from the conductance hay moisture meter which was field tested by the Dairy Science Departments, University of Maryland, and Texas A&M University, in 1965. Several instrument manufacturers have indicated an interest in manufacturing the meter for commercial sale. Over 500 requests for copies of USDA ARS 51-2 were filled during the past two years which indicates the amount of public interest in the moisture meter.

5. Development of Seed Germinator. Tests with the new germinator showed several minor modifications to be necessary. A programmed temperature control for the coolant liquid was designed to prevent overshoot in cooling. Improvement was made in light uniformity and intensity levels over commercial germinators presently in use. Humidity and temperature control appear to be excellent based on limited tests of the new germinator.

6. Processing Grass Seed for Laboratory Testing. Extensive tests made with a modified McGill rice sheller to dehull the caryopses of sideoats grama and orchardgrass showed that the mechanical method (1) measures the purity with somewhat higher sensitivity than the official method; (2) is twice as fast as the hand method; (3) does not require a highly trained seed analyst, and (4) provides shelled seed that germinate in one-half to two-thirds the time required for unshelled seed. Limited tests show the method is equally well suited for big bluestem, little bluestem, and blue grama.

7. Seed Vigor. Respiration measurements were highly correlated with the vigor of corn seed which had been stored under various conditions of temperature and humidity for four years. Respiration also detected weak lots of cabbage, onion, and tomato seeds; and revealed desiccation-induced dormancy in sorghum. Calculation of R. Q. (carbon dioxide evolution/oxygen

uptake) ratios and the use of an oxygen atmosphere during the respiration tests sometimes revealed weak seed lots more effectively than simple determinations of oxygen uptake, especially in tests performed during the initial 2-4 hours of germination. Arrangements were made with the Seed Branch of the Grain Division, Consumer and Marketing Service, for conducting practical tests of seed vigor by the respiration method.

8. Verification of Varietal Designations of Crop Seeds. Electrophoretic analysis of protein extracts from 65 soybean varieties revealed an outstanding difference among the varieties, based on two banding patterns, tentatively called A and B. Each variety consistently contained proteins which produced either an A or B banding pattern. F₁ progeny of crosses between A and B type varieties yielded both A and B proteins. Pubescence color of soybean seedlings was intensified and more easily detected when continuous high-intensity light was used in a growth chamber.

Extracts from perennial ryegrass seed contained a distinctive protein not found in annual ryegrass seed. It is possible that this protein difference may form the basis for a practical test to distinguish between seed of the two species.

9. Detection and Identification of Seedborne Pathogenic Fungi. The influence of 128 culture media, consisting of a basal Czapek's medium to which were added different combinations of 32 carbon-containing compounds and four nitrogen-containing compounds, on the growth of 30 seedborne parasitic and saprophytic fungi were examined. Although a final evaluation of the data will not be possible until a statistical analysis has been completed, it is apparent that no selective action was obtained with any of the culture media used.

10. Environmental Control of Seed Dormancy and Germination. Seeds of two ecotypes of Eragrostis curvula were similar in their germination responses. Both germinated well in continuous light following a dark imbibition period and less well when the dark period was interrupted briefly by red light. Phytochrome control was clearly evident because far red irradiation completely nullified the inhibitory effects of red. The two ecotypes differed as follows when exposed to 29°C.: (a) Lot 1 seeds germinated when placed in continuous light, while Lot 2 seeds required a long dark period; (b) after dark-treatment, Lot 1 seeds responded to a brief exposure to red light while Lot 2 seeds still required a long period of continuous or intermittent light for germination. These results emphasize their importance of small variations in light and temperature for successful germination of light sensitive seeds.

11. Germination Inhibitors in Seeds. Extracts of barley seeds yielded the following compounds which were identified and found to inhibit the germination of lettuce seeds: P-hydroxybenzoic acid, P-coumaric acid, P-hydroxybenzaldehyde, and vanillin. At least five other acids of phenolic character were found, but these have not been identified.

12. Biological and Environmental Factors Affecting the Physiological Maturity of Grass Seeds. The first six-month report indicates that seeds of orchardgrass and annual ryegrass harvested in the dough stage gave higher germinations than when harvested in full ripe stage. Seeds of meadow fescue and Kentucky bluegrass did not exhibit this phenomenon. All results are tentative.
 13. Development of Methods Suitable for Routine Testing of Seed for Seedborne Organisms. In previous research under this grant, significantly fewer seed-borne fungi developed in germination tests when the blotters were kept wet than when kept slightly dry. Research during the past year showed that this effect is due to an antagonistic effect of bacteria. A screening test for seedborne fungi on wheat grown in the Netherlands showed that Fusarium nivale was the most prevalent pathogen; however, the less prevalent fungi, F. graminearum and Septoria nodorum, appeared to be more serious to the growing crop.
 14. Moisture Levels for Seed Germination. Seeds of a number of species germinated equally well over a range of moisture levels; thus, no specific amount of moisture was found to be optimum for germination of seeds of the species used in this study. Using a standard experimental procedure, the following kinds germinated equally well over the indicated range of moisture levels (i.e., number of ml of water added to 400 g of uniform sand): Peanut, 70-85 ml; sorghum, 15-55 ml; watermelon, 30-80 ml. The speed of germination of rice was retarded by the addition of excess water during the first three days of test; however, the total germination was not affected when the seedlings were evaluated 10-14 days after planting. Addition of excess water after three days from planting date had no influence on the speed of germination.
- B. Quality Maintenance in Storage
1. Microbiological Deterioration of Grass Seeds During Marketing. Storage of four species of grass seeds under 16 different levels of temperature and relative humidity revealed that freshly harvested seeds of Kentucky bluegrass, red fescue, tall fescue, and annual ryegrass apparently harbor storage fungi that are in a dormant state. Plating-out on agar seeds of these grasses before and after storage for three months at 30°C. and 75 percent relative humidity revealed no storage fungi prior to storage, but numerous storage fungi after the storage period. To separate the effects of processes inherent in seeds from the effects of microorganisms while studying the effects of storage environment on seed germinability, it is necessary to use seeds free of storage fungi. Since it is not possible to obtain grass seeds from the field free of storage fungi, efforts are being made to produce a supply of germ-free seeds.

PUBLICATIONS REPORTING RESULTS OF USDA AND COOPERATIVE RESEARCH

Objective Measurement and Evaluation of Quality

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PUBLICATIONS -- STATE EXPERIMENT STATIONS AND COOPERATIVE PROGRAMS

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MARKETING FACILITIES, EQUIPMENT AND METHODS
Transportation and Facilities Research Division, ARS

Problem. Differences in varieties of individual field crops and in the environments of producing areas where they are conditioned and stored, together with advancing techniques in cultural and harvesting practices, require new or modified marketing facilities, equipment, and methods. Such changes are essential to the efficient and economical handling, conditioning, and storing of these crops and to maintaining their quality. There is a need for improved designs for facilities based on functional and structural requirements, which will expedite the movement of commodities into, within, and out of the facility. There is also a need for handling and conditioning equipment which will minimize labor and other costs and also minimize the extent of physical damage (breakage) to the grain as it is handled into, within, and out of marketing facilities. More knowledge is needed of the relative efficiency of various handling and conditioning methods so that improved or revised methods and equipment can be developed to perform necessary operations.

USDA AND COOPERATIVE PROGRAM

The Department has a long-term program involving engineers engaged in both applied and basic research on, as well as application of known principles to, the solution of problems of handling, storing, and conditioning field crops in marketing channels. Grain aeration and drying research is carried out at Manhattan, Kans., on wheat and grain sorghum and at Lafayette, Ind., on corn, in both laboratory and pilot-scale facilities and in commercial storages; in cooperation with the Agricultural Experiment Stations of respectively Kansas and Purdue University (Indiana), the Market Quality Research Division, and with grain storage firms; and is supplemented by a research grant by the Corn Industries Research Foundation made through Purdue University. Investigations designed to determine the extent and causes of physical damage (breakage) to grain by handling equipment used in marketing facilities are conducted at Manhattan, Kans., in cooperation with the Kansas Station; and are supplemented by a research contract with Cargill, Inc., Minneapolis, Minn. Investigations on the basic theory of dynamic and static pressure phenomena in grain under storage conditions, by theoretical analysis and simulation, are conducted under a research contract with Dr. J. D. Isaacson, Adelphi, Md. Studies to develop improved techniques and equipment for uniformly blending heterogeneous lots of seed are conducted under a research cooperative agreement with the Mississippi Agricultural Experiment Station, the Consumer and

Marketing Service, and the Field Seed Institute of North America. Studies on the handling, drying, aerating, and storing of rice are conducted at Beaumont, Tex., in cooperation with the Texas Agricultural Experiment Station and at commercial facilities in Texas, Arkansas, Louisiana, and Mississippi.

The Federal effort devoted to research in this area during the Fiscal Year 1966 totaled 10.6 scientific man-years, including 5.3 man-years for contracts and research cooperative agreements: 2.3 to grain aeration and drying; 4.1 to the effect of handling equipment and methods on extent of physical damage to grain; 1.0 to the handling, drying, aerating, and storing of rice; 1.2 to grain transport equipment; and 2.0 to program leadership.

REPORT OF PROGRESS FOR USDA AND COOPERATIVE PROGRAMS

A. Effect of Handling Equipment on Physical Damage to Grain

1. At Minneapolis, Minn., tests are underway under the research contract with Cargill, Inc. to determine the extent and causes of physical damage (breakage) to grain by handling equipment used in commercial facilities. All equipment required for conducting the specified tests on corn and wheat has been installed and is in use. Some 250 grain thrower, grain drop, and bucket elevator test runs have been completed using spring and winter wheat at two different moisture levels and at low temperatures between 33° to 50° F. The following is a summary of the results of the breakage tests completed:

<u>Test Treatment</u>	<u>Wheat</u>	<u>Wheat moisture percent</u>	<u>Maximum Percent of breakage</u>
Thrower	Winter	10.5-11.5	0.22
Thrower	Spring	10.5-11.5	.30 <u>1/</u>
Thrower	Spring	12.5-13.5	.27
Drop (free fall)	Winter	10.5-11.5	.30 <u>2/</u>
Bucket elevator	Winter	10.5-11.5	.27 <u>3/</u>

1/ Belt speed, 4,030 f.p.m.; bulkhead at 10-foot distance; grain temp. 36° F.

2/ 100-foot free fall onto concrete; grain temp. 44° F.

3/ Bucket speed, 940 f.p.m.; back feeding; grain temp. 49° F.

During free fall drop tests the grain stream assumed the following shapes at a few feet below the discharge office: (1) Approximately a 6-inch diameter irregular circle from a 12-inch diameter orifice; and (2) approximately a 4- by 6-inch oblong shaped stream from an 8-inch diameter orifice. With free

fall of grain on grain, the maximum penetration of the grain stream into the static grain was approximately 18 inches after a 100-foot drop. In bucket elevator tests with a belt speed of 940 f.p.m. (and no head cover over the top of the elevator) the trajectory of the grain leaving the buckets reached a maximum height of 8 feet above the head pulley shaft.

2. At Manhattan, Kans., a new laboratory was set up under a cooperative agreement with the Kansas Agricultural Experiment Station to investigate design principles and operating methods for handling equipment that affect the extent of physical damage to grain. These basic studies supplement the simulated full-scale tests being conducted by Cargill, Inc. under a research contract. Reports have been reviewed of past and current work concerned with physical damage to grain and of work concerned with various properties of grain, such as shape factor, porosity, terminal velocity, elasticity, resilience, breaking strength, etc. Possible techniques, equipment needed, and theoretical background for testing and evaluating grain breakage were evaluated. Various instruments and equipment needed for the studies have been installed in the new laboratory.

In order to know the initial physical condition of the grain used in the Cargill tests, samples are obtained from each lot of grain prior to testing and these samples are sent to the Manhattan laboratory for analysis. Samples are analyzed for susceptibility to breakage, percent of kernel cracks, and for percent viability; also, for physical properties such as bulk density, porosity, average weight, volume of kernel, etc. Duplicate samples are also sent to the Standardization and Testing Branch, Grain Division, C&MS, Beltsville, Md., for studies on susceptibility to breakage.

Limited tests have been made to determine the effect of repeated wetting and drying cycles on the internal cracking of grain. Samples of winter wheat were wetted to approximately 20 percent moisture; then dried to about 12 percent. These cycles were repeated three times each at drying air temperatures of 100°, 120°, 140°, 160°, 180°, and 200° F. Samples at each stage were examined radiographically for internal cracks and tested by breakage tester for susceptibility to breakage. Generally, there was some increase in breakage during each successive cycle regardless of drying temperature. As expected the largest increase in breakage occurred at the higher drying temperatures. For example, at 100° F. the percent breakage amounted to 0.55 to 0.60; at 200° F. it amounted to 1.8 percent.

Similar experiments were conducted with white corn at drying temperatures of 120° and 180° F. The amount of breakage was much greater than for wheat. For example, at 120° F., the percent breakage was 3.0, 6.0, and 8.9 respectively for each successive cycle. At 180° F., the percent breakage was 7.5, 8.5, and 16.5.

B. Grain Aeration and Drying

1. Aeration. At Manhattan, Kans., field studies were continued on a limited basis in commercial flat storages. At Hastings, Nebr., grain sorghum was carried over for a fourth winter in a large steel flat storage building, 100 by 360 feet. This same lot of grain has been in this storage for nearly four years. During the two previous winters serious caking and spoilage occurred in the peaked-center surface area due to high temperatures and moisture transfer. The importance and advantages of aeration during the early winter months in these large storages was demonstrated during the past winter. Aeration fans were operated during cool weather in October and November until grain temperatures were between 30° and 53° F. Surface samples submitted for official grade indicated no additional deterioration had taken place nor was there any appreciable amount of moisture migration when checked in December.

Research to develop and test crossflow ventilation systems for deep silo-type storages was continued in 1965-66 with wheat and grain sorghum. At Abilene, Kans., the crossflow test bin was filled and used for three different 25,000-bushel lots of tough new wheat. The first lot had an initial moisture content of 14.1 percent before ventilation and 13.3 percent, afterwards; and the calculated amount of moisture evaporated was 11,400 pounds. The fans were operated 124 hours during daylight working hours. The initial cooling front moved through 16 feet of wheat at the bin center (diameter) in 22 hours, while the longer distance through grain near the bin walls required 30 hours. The second lot contained 14.0 percent moisture and fans were operated continuously for 8 days to obtain 178 hours of ventilation. The calculated amount of moisture evaporated was 16,200 pounds. The initial average grain temperature was 91° F. and the final average was 73° F., a reduction of 18° F. The third lot of tough wheat was sampled into the crossflow bin on July 31 for cooling and storage until transferred to the flour mill. The grain had an average initial temperature of 87° F. and was cooled to an average temperature of 76.5° F., using 88 hours of fan operation.

The crossflow system was also used for four different lots of new crop grain sorghum which were too wet for storage without aeration or drying. An average of nearly 370 pounds of water per hour were removed during the first 24 hours of fan operation compared with only 25 pounds per hour after 60 hours of operation when the grain temperature was near the air temperature.

A manuscript reporting the results of the cross flow ventilation tests conducted to date has been prepared and submitted for publication.

In a continuing study, grain temperature data were again obtained in a concrete upright storage, 18 feet in diameter and 110 feet high, at Cheney, Kans., to determine the effect of ambient air temperatures on the temperature of wheat in static (unturned) storage. The 1964-65 data provide additional needed information on the cyclical effect of seasonal temperatures on stored wheat and supplement data previously obtained from this same bin of wheat.

The annual cycle-change in grain temperatures indicates the variations in heat transfer conditions depending upon the season or month of the year. The maximum spread in grain temperatures occurred in January 1966 with wheat at 22° F., six inches from the wall and at 62° F., near the bin center which is about 10 feet from the wall. The reverse of the cycle occurred in July when the temperature of the grain near the outer wall was 83° F. and grain near the bin center at 55° F.

2. Drying. At Lafayette, Ind., further tests on the aeration-cooling portion of the dryeration process for drying field-shelled corn, showed that increasing the length of the tempering period between heated-air drying and aeration-cooling increased the number of sound kernels having no stress cracks. This increase in sound kernels was greatest when the tempering period was advanced from 2 to 8 hours and smallest when advanced from 8 to 12 hours. Slow cooling after the tempering period was also beneficial. However, the effect of these different tempering periods and cooling speeds on the wet milling properties of the dried corn showed an opposite trend. While the "millability scores" of samples from all tests were in the acceptable range, the best scores were with the 2-hour tempering period and the faster cooling rates tested. The maximum moisture removal from the corn occurred using a tempering period of 8 hours followed by 10 hours of aeration-cooling with an airflow rate of 0.5 c.f.m. per bushel. These results confirmed previous recommendations for the tempering period and the cooling airflow rate. A test, conducted to determine if corn could be tempered and stored in the same bin where aeration-cooling was performed, showed that corn could be successfully stored when cooling was completed within 24 hours if aeration was continued during favorable weather throughout the storage period.

The storage of 18 percent moisture corn in insulated bins equipped for aeration-cooling was evaluated in terms of the potential hazard of toxin-producing mold growth. Aeration was controlled in 3 bins in an attempt to maintain stored corn temperatures of 50°, and 40°, and 30° F. The fourth bin of corn was aerated continuously with an airflow rate of 0.6 c.f.m. per bushel, the same as used in the other bins. All test lots were stored from early November to the following May without damage or evidence of mold growth, except for corn in the bin held at 50° F. Here the corn near the surface showed increases in both Penicillium ssp and A. glaucus in the last month of storage.

Eight drying tests were performed in the laboratory concurrent-flow dryer. Both the initial drying air and the corn temperatures changed rapidly (approaching each other), then both gradually decreased through the remaining depth of the dryer. Corn of 33 percent moisture was dried to 13.6 percent, with hourly drying capacities ranging from 0.58 (200° F. air at 50 c.f.m. per square foot) to 2.00 (350° F. air at 90 c.f.m. per square foot) bushels per square foot of bed area. About 1/2 percent moisture was removed following drying using a counter-flow cooler. Replacing the cooler with dryeration

resulted in removing 1.1 percent of moisture. Stress cracks and milling quality evaluations showed that using the cooler after drying did not reduce brittleness. Millability scores decreased with increased drying temperatures--at 300° F. the score was 79.4, slightly short of an acceptable score of 80.

Three drying tests were performed in the counter-flow dryer. Unlike the concurrent-flow dryer, here the drying air moved in a direction opposite to the flow of corn, exhausting at a temperature only slightly above the initial corn temperature. The corn was discharged at a temperature near that of the initial drying air. The heat absorbed by the corn through the dryer depth resulted in a high drying efficiency and made dryeration an efficient method for further removal of moisture. Corn of 22 percent moisture was dried to 13 percent with hourly drying capacities ranging from 1.2 (200° F. air) to 1.9 (300° F. air) bushels per square foot of bed area. Quality evaluation of the corn indicates this method of drying is more detrimental than concurrent flow.

Performance of grain metering devices for continuous grain dryers was further evaluated. Observation of corn as it was removed from a model grain column using a conventional fluted-roll device showed that a 1/2" clearance between the roll and housing is best for a positive grain discharge with a minimum amount of kernel breakage. Preliminary tests indicate that the amount the roll is exposed to the hopper outlet has a marked effect on capacity. Neither the height of the corn column nor direction of roll rotation showed a significant effect on capacity. However, increasing the roll speed five-fold (0.9 to 4.6 r.p.m.) increased the corn throughput only 4.8 times.

C. Handling, Drying, Aerating, and Storing Rice

1. Drying. At Beaumont, Tex., tests using a pilot-size continuous-flow heated-air dryer were conducted to determine if, following passes through the dryer, tempering of binned rice prior to cooling by aeration resulted in removing more moisture or in better milling quality. Results showed that, (1) tempering rice for periods up to 12 hours before cooling did not change the milling quality or significantly increase the amount of moisture removal when the rice was later cooled and (2) in order to save storage time between dryer passes the recommended procedure would be to start aeration immediately after rice from the dryer is loaded into a bin. From the test results it appears that tempering rice for as long as 12 hours at the rice moistures (up to 20 percent) and rice temperatures (110° to 145° F.) used will not create any detectable production of aflatoxins. Both laboratory and full-scale tests showed that certain varieties of rice respond differently to various drying procedures. At any heated air temperature used (113° to 172° F.), TP 49 (long-grain) dried faster than either Nato (medium-grain) or Belle Patna (long-grain). Belle Patna dried faster than Nato when air temperatures up to 146° F. were used, but at higher temperatures Nato dried as readily as Belle Patna.

2. Aeration and Storage. At Beaumont, Tex., the study on the use of aeration for maintaining quality of undried rice was continued. During the 1965 harvest season, three batches of green rice were stored and sampled periodically for determining rice grade. This series of tests was initiated in 1961 and data have been obtained on 15 batches of rice stored at initial moisture contents in the range between 18 and 24 percent (wet basis) and with the average ambient air temperature ranging between 66° and 85° F. Airflow rates of 0.5, 1.0, and 2.0 c.f.m. per barrel were replicated three times. Information from these tests has been used in preparing charts for use in predicting the safe allowable storage time for green rice under varying rice moistures and outdoor temperatures. In cooperation with the College Station, Tex., office of the Market Quality Research Division, samples of rice from the three batches stored during the 1965 harvest season were assayed for aflatoxins. Detectable amounts of aflatoxin were found in rice stored with an initial moisture content of approximately 22 percent when the outdoor temperature averaged approximately 82° F. None was detected in rice stored at 20 percent moisture content when outdoor temperatures averaged approximately 70° F.

3. Handling of Rough Rice at Commercial Dryers. Handling of rice during drying operations was studied at four commercial rice dryers in Texas and Louisiana during the 1965 harvest season. These studies showed that unproductive time resulting from wait time during filling of the dryer can be reduced by installing a large garner bin above each drying unit. When switching from one lot of rice to another, the garner bin can be filled with undried rice while dried rice is emptying from the dryer. Rice will flow from the garner bin into the dryer at a fast rate and approximately 25 minutes per pass per lot can be saved if the garner bin is at least as large as the associated drying unit. Time studies of labor inputs for drying rice indicated that a dryer having a good plant layout, where one operator handles four dryer units, used 3.7 man-hours per 100 barrels of rice dried as compared with nearly 6 man-hours per 100 barrels dried at an installation having a poor layout where three operators are required for three drying units. The electric power requirements at one location were 30 kilowatt-hours per 100 barrel-lot requiring five dryer passes. The overall power requirements for the entire plant, including aeration, receiving, etc., was 180 kilowatt-hours per 100 barrels; which indicates that although aeration required several times as much power as dryer operation, drying capacity was increased by about 13 percent as fewer dryer passes were required.

4. Bulk Handling of Milled Rice. Tests were continued to determine the amount of breakage which results when milled rice was dropped into bins from different heights. Milled rice at 11 percent moisture content was more susceptible to breakage than the same variety at 13 percent moisture content when both were dropped 30 feet onto a flat steel surface. Another series of tests failed to show a difference in the amount of breakage when milled rice at two

different temperatures; 30 and 80° F., but having the same properties otherwise; was dropped from 30 feet onto a steel surface. Both brown rice and par-boiled rice were found to be less susceptible to breakage under similar test conditions than well milled rice.

D. Uniform Blending of Seed

At State College, Miss., under a research cooperative agreement with Mississippi Agricultural Experiment Station, a study was initiated to more uniformly blend seed of heterogeneous lots. Variations in model blending bins will be constructed and tested for their effectiveness with cereal, forage legume, and forage grass seeds. The most efficient bin will be selected for further full-scale studies, then plans and specifications developed for the best blending bin.

E. Grain Transport in Boxcars. The field survey portion of a study to determine amounts of grain losses from various factors during transport in boxcars was completed. This field survey was done under contract with an outside research organization. Visual inspection was made of 1960 cars of wheat, corn, and soybeans arriving at terminal elevators in Kansas City, Missouri; Minneapolis, Minnesota; and Chicago, Illinois.

At the end of FY 66 data from the field survey was being placed on punch cards for machine processing for statistical analysis. Preliminary study of the data indicates that, out of 1960 cars inspected, only 370 or 19 percent were clear, i.e., without defects. Most of the clear cars were either new, rebuilt or reconditioned. Defective grain doors were found in 260, or 13 percent of the cars. The results suggest that it would be worthwhile to develop a new type grain door that would be more effective in preventing grain leakage and which would reduce damage to the cars.

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COOPERATIVE MARKETING
Marketing Division, FCS

Problem: Farmers continue to expand their use of cooperatives in marketing the products of their farms. In light of the rapid and complex changes taking place in technology and in market organization and practices, research is needed to help farmer cooperatives and other marketing agencies perform needed marketing services both more efficiently and more effectively. Farmer-directors, managers and others, including the public, need more information to assist in making decisions on how cooperatives can maintain and strengthen the bargaining power of farmers, increase efficiency and reduce costs of marketing, and better meet the needs of our mass distribution system for large quantities of products on a specification basis.

Farmer cooperatives are an important part of the distribution system and represent a major potential for meeting farmers' marketing problems in our modern, dynamic system. They are organized and operated to increase farmers' net income. However, cooperatives face many problems in achieving this goal. Cooperatives must find ways to consolidate volume, for example, through internal growth, merger, acquisition or federation, to strengthen their market position and meet the needs of mass merchandising. Ways must be found to reduce costs by increasing efficiency through improved operating methods, better organization and management, and more use of new technologies.

USDA AND COOPERATIVE PROGRAM

The Department conducts a continuing long-range program of research and technical assistance on problems of marketing farm products cooperatively. Studies are made on the organization, operation, and role of farmer cooperatives in marketing. While most of the research is done directly with cooperatives, the results are generally of benefit to other marketing firms. The work is centered in Washington, D. C. Many of the studies, however, are done in cooperation with various State experiment stations, extension services, and departments of agriculture.

The number of Federal scientific man-years devoted to this research totals 3.0, of which 1.8 man-years relate to work on grain, 0.1 on rice, and 1.1 on feed.

PROGRAM OF STATE EXPERIMENT STATIONS

Most commodity marketing research of the agricultural experiment stations is helpful to marketing cooperatives. Some projects, however, deal specifically with cooperative marketing problems, opportunities, and impacts. The total research effort on cooperative marketing in the State experiment

stations is 0.8 scientific man-years.

Many State experiment stations do a considerable amount of research on technical aspects of feeds, fertilizers, seed, farm equipment, and farm supplies that is of value to purchasing cooperatives. A limited number of projects also deal specifically with cooperative purchasing problems, opportunities, and impacts.

The total research effort on cooperative purchasing in the State experiment stations is 0.8 scientific man-years.

A large part of the research in this area is devoted to the financing problems of cooperatives. This involves study of the financial structure and financing methods of cooperatives to determine those that are the most satisfactory from the standpoint of providing adequate capital for growth and treating patrons equitably.

Other research deals with cooperative information sources in performing management functions of planning and controlling, membership control of cooperatives, and socio-economic relationships existing between low-income farmers and the cooperatives to which they belong.

A total of 2.1 scientific man-years is being devoted to research in this area.

REPORT OF PROGRESS FOR USDA AND COOPERATIVE PROGRAMS

A. Potentials in Cooperative Marketing

The present and potential role of cooperative marketing requires study in several commodity areas. Current information on cooperative operations can be related to production and marketing conditions.

The objective of such research is to develop recommendations about operations and services of existing cooperatives and particularly to identify opportunities for farmers to increase their marketing returns by developing significant new areas of cooperative operation.

Grain. With increasing quantities of wheat and flour being purchased on specific quality characteristics and with increasing competition in merchandising, several of the regional grain cooperatives have installed experimental milling and baking laboratories so they can determine the quality of wheat received and, in turn meet the specifications of the flour miller. A study of the use, effectiveness, costs and benefits of these laboratories is just getting underway.

B. Improving Operating and Handling Methods

Research was underway in several commodity fields to examine new methods, equipment, and structures of efficient and safe processing and storage of agricultural products by cooperatives.

Rice. A study of costs and other economic considerations in drying and storing rough rice in 74 on-farm and 21 off-farm facilities was completed. Among on-farm units, buildings with bulk bins incurred the highest operating costs followed by round bulk bins and multipass units. For the off-farm commercial facilities, no significant difference existed in the total combined cost of drying and storing between cooperative units and those not cooperatively owned. Little average differences existed in the quality of rice resulting from the use of on-farm and off-farm facilities.

C. Improving the Organization, Financing, and Management of Marketing Cooperatives

Studies were made to determine ways to improve the efficiency and assist cooperatives improve their services by analysis of organization, financing, and management practices.

Grain. A study of inventory controls, practices, and responsibilities at local cooperative elevators was completed. A report is being prepared describing the wide variety of practices and methods used to control grain inventories in local elevators. The annual analysis of the operations and financial status of regional grain cooperatives was continued. These cooperatives are handling increased volumes of grain and with increased emphasis on merchandising, both domestic and export. Work is underway to determine the feasibility of establishing cooperative deep water elevators at Southeast and Gulf Coast ports. Work continues on studies of grain marketing, processing, utilization, and transportation problems that are peculiar to the South.

D. Improving Sales and Distribution Methods

Purchasing cooperatives constantly need to be studying ways to increase volume to lower per unit handling costs, methods for developing more effective systems of distributing supplies and providing services to rural users, adjustments to changes in amount and type of supplies and services required by large farm operators, and improved methods of serving rural residents where suburban and industrial developments are expanding.

Feed. A report covering the study of 16 feed ingredient and supply buying groups in Illinois was completed. Principal findings were reported last year.

Another study of the organizational and operating features of 16 poultry feed bargaining groups in California was completed. Each group bargained with mills to manufacture specified formulas for their members. Principal findings were as follows: (1) Sixteen groups, formed from 1948 to 1963, had from 2 to 35 members. The feed requirements of the groups ranged from 1,380 to 27,100 tons a year with the median of about 7,200 tons. Members had flocks below average in size. (2) It appeared the groups had been successful in lowering feed prices to the minimum possible within the framework of the existing industry, and yet had maintained the quality of feed. (3) Based on limited information, members of some groups were getting formula feeds delivered to their ranches for as low as \$4.60 to \$5 a ton above weekly ingredient costs, with payment due 10 to 14 days after delivery date. (4) Indications were that individual poultrymen with flocks of more than 40,000 layers can obtain all the price concessions given to bargaining groups. (5) With the trend to larger flocks and more farm mills in California, small milling firms may have to increasingly depend on poultrymen with small flocks, most of whom will be members of some group.

Most of the field data were obtained for a study of the practices cooperatives employ in pricing feed at the wholesale and retail levels. Special attention is being given to various pricing schedules based upon types of quantity and volume discounts, cash discounts, and charges made for various services.

Preliminary findings indicate that among cooperatives in the Midwest and Eastern United States, considerable variation exists in policies and practices in pricing feed. Some regional wholesale cooperatives give quantity or volume discounts to their retail outlets, and most grant price adjustments or concessions to enable locals to meet competitive situations.

Approximately one-half of the retail outlets grant patrons a quantity discount often ranging from \$1 to \$3 a ton on individual feed orders of specified tonnages. Less than half the retail outlets grant patrons a volume discount of from \$1 to \$2 a ton when feed purchases over a given period of time reached a certain volume. Only a few of the retail outlets grant patrons both types of discounts. The amount of discounts is greatest in areas where there is the greatest disparity in the volumes purchased by individual farm operators.

E. Potentials of Cooperatives in Farm Production Supplies and Farm Services

Studies were made of the present status and the possibilities of cooperatives procuring supplies and services more effectively for farmers and other rural residents. This includes diversification, integration, expansion in present operations and areas, and expansion into new areas and services. Research will aid directors and managers in planning and implementing programs.

Feed. A study was initiated through a contract with a State university to identify the factors and develop guidelines for determining the feasibility of establishing alfalfa dehydration plants.

F. Improving Operating and Handling Methods for Farm Production Supplies

Studies were made to assist cooperatives in providing farmers with needed supplies and services in the least cost manner and by methods that save them time and labor on the farm.

1. Feed. Work on a study of the type, cost, operating expenses, problems, and benefits of bulk feed relay or transfer stations was deferred because of loss of key personnel.

2. Seed. Work on a study of the feasibility of bulk receiving and distribution of seed, including methods, costs, problems, and benefits, was deferred.

G. Transportation and Physical Distribution by Farmer Cooperatives

1. Loss and Damage to Agricultural Products in Transit. A report of findings was completed on a study of weight losses on 13,611 rail shipments of grain from 107 elevators and 777 barge shipments of grain from river facilities of six regional grain marketing cooperatives. The study showed that losses during handling and transportation amounted to 558 pounds of grain per car shipped. Of 700 rail cars furnished to elevators for loading, 85 percent were found to have defects that could contribute to losses. The study also showed that less than half of the 107 elevator managers filed claims for recovery of losses although all experienced such losses.

2. Motortruck Operations and Costs. A report of findings on a study of costs and practices of cooperatives in operating bulk-feed trucks was published. Principal findings of the study are: (1) Total truck operating costs per ton of feed delivered ranged from \$2.39 to a high of \$7.09 and averaged \$2.89. (2) Truck operating costs, less administrative expenses, averaged 48 cents a mile operated. The report recommended that detailed truck cost and performance records be maintained on individual trucks rather than on a fleet basis only.

3. Grain Transportation and Handling by Farmer Cooperatives. Preliminary findings of a study of ownership and leasing of covered hopper cars by cooperative include: (1) The major reason cooperatives operate their own hopper cars was to help alleviate a chronic shortage of boxcars. (2) The 5½ cents a mile loaded and empty allowance paid by the railroads for use of shipper owned or leased hopper cars contributes to wasteful transportation. The allowance should apply on loaded miles only as this would

encourage the shipper to seek return loads in his equipment. (3) There are opportunities for shippers to reduce costs by owning rather than leasing hopper cars.

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ECONOMICS OF MARKETING
Marketing Economics Division, ERS

Problem: Economic research in agricultural marketing revolves around the problems of increasing efficiency in the processing and distribution system and providing a foundation for orderly adjustments to changes inside and outside of agriculture. Marketing must be looked upon as a dynamic and changing process. The capacity to adjust to and cope with the dynamics of modern marketing is required increasingly of producers and distributors of farm products. Demands of a more knowledgeable and sophisticated consuming public are adding to the pressures for an even more rapid escalation of developments and changes within the marketing system. Changes in institutions and redirection of public policies and programs are modifying the economic environment in which marketing firms must perform and operate. Because of rapid changes and increasing complexities associated with a dynamic marketing system, it is necessary that a continuous program of research be conducted in marketing--a program aimed at keeping producers and marketing firms abreast of the flow of events and providing information necessary to them in making proper and orderly adjustments to change.

Of increasing economic concern is the problem of how to improve and strengthen markets for farm products in face of a continuing rise in production, higher distribution costs, and competition from nonagricultural products. The problem of increasing demand for farm products to meet rising productivity has become progressively more pronounced in the last decade. Interest in the development of markets has mounted as larger and larger financial outlays become necessary for price-support operation and maintenance of reasonable levels of farm income.

Through research on the different facets of market development, information is provided farm groups, processors, and distributors which enables them to make the most of new market outlets and opportunities. In addition, they are better able to assess the demand of consumers from the standpoint of kinds and forms of food and services deemed most satisfactory.

USDA AND COOPERATIVE PROGRAM

The Department has a continuing long-term program of economic research relating to grain and forage crops. The research involves economists and personnel with dual economic and technical training and is carried out at Washington, D. C., and field offices in Albany, California; and Peoria, Illinois, and in cooperation with State Agricultural Experiment Stations in a number of States. The grain and forage phases of the research involve 13.9 Federal scientific man-years of which 4.5 are related to the physical efficiency of performing marketing functions; 3.0 to the orientation of input and output of marketing firms; 2.1 to the orientation of consumers in marketing and 4.3 to location economics.

PROGRESS -- USDA AND COOPERATIVE PROGRAMS

A. Prices, Margins, and Costs

Major emphasis on grain products price spreads during the past year has been on assisting the National Commission on Food Marketing staff in planning and conducting an analysis of the milling and baking industries. Data sources developed during this work are being consulted in an effort to obtain similar information on a continuing basis. In addition, a new retail price reporting system for bread is being tested. The marketing spread for a 1-pound loaf of white bread was 18.8 cents in August 1966, 1.4 cents above the level of a year earlier. The farm value of ingredients showed a similar rise from 3.4 to 4.0 cents, the sharpest increase since 1947 to the highest level since January 1948. Data from several sources are being analyzed with the objective of revising the series on these spreads, and also of improving the source of information used in measuring the breakdown of the farm-retail spread among the various segments in the marketing process.

A national study was completed which examined the costs of storing and handling grain in commercial elevators for 1964-65. Four types of cost were analyzed: (1) out-of-pocket costs, which contained no allowance for interest on investment or depreciation; (2) total book costs, which included the firms' own figures for depreciation and interest on investment; (3) short-term competitive rates reflecting the out-of-pocket cost of the marginal firms; and (4) long-term competitive rates based on total costs including depreciation and interest on the replacement value of capital assets. Country elevator records showed book costs for storing and handling grain by the most common method averaged 10.4 cents per bushel and ranged from 9.2 cents in the South and East to 12.0 cents in the Great Lakes area. This cost included 1 year's storage plus receiving by truck and shipping by rail. Short-run competitive rates for the combined storage and handling function averaged 8.0 cents per bushel. Long-run competitive rates averaged 13.5 cents per bushel. At inland and port terminals, the combined book cost for receiving, storing, and shipping averaged 9.0 cents and 9.3 cents per bushel, respectively. Short-run competitive rates were 7.0 cents per bushel. Long-run competitive rates averaged 12.0 cents at inland and 13.5 cents per bushel at port terminals. Storage cost alone (as shown on the warehouse books) averaged 5.4, 5.5, and 7.2 cents per bushel at country, inland, and port terminals.

Economic and technological changes in the mixed feeds industry during recent years have been quite pronounced and widespread. Feed manufacturing operation is one of the greatest areas of change. Research had made use of economic-engineering models to show management where efficiencies are possible. Four models, ranging in size from 80 tons to 260 tons per 8-hour day, are being analyzed. Labor requirements range from 0.80 man-hours per ton to 0.53 man-hours per ton. The impact of product specialization is receiving special attention as a factor in obtaining a low-cost and high efficiency plant operation.

Substantial economics to scale exist in the manufacturing of pelleted broiler feeds over a range in mill capacities from 20 to 348 tons a day, but the relatively high cost of bulk feed distribution tends to offset these economies except in situations where broiler production is extremely dense and concentrated about the mill site.

Investment for the seven model mills for manufacturing layer feed, which range in capacity from 65 to 800 tons a shift (8 hours), increases from approximately \$200,000 to \$1,167,000. Preliminary indications are that fairly substantial economies probably exist over a portion of this range in mill sizes.

Mash poultry feed in bulk form for layers can be manufactured for approximately \$2.44 a ton, with operations at 415 tons capacity for two full shifts a day. Bagging in 50 pound paper adds approximately \$4.00 a ton.

Present alfalfa hay grades do not reflect some quality attributes. Thus efficient price determination is difficult. A study in cooperation with the Nevada Agricultural Experiment Station involved developing experimental alfalfa grades which accurately reflect quality factors. The experimental grades retain most of the present U. S. hay standards but add criteria and test methods for protein and moisture. A market test of these grades showed that the experimental grades provided sufficient information for their use in price determination; and they will be used with increasing frequency, either as amended U. S. hay standards or as informal agreements among the trade.

B. Location and Interregional Competition

Transportation

Intermodal competition for whole-grain traffic is continuing. Railroads are now attempting to adjust rates and services so as to hold their volume and in many instances are making determined efforts to regain traffic lost to barges and trucks. The numerous studies started over the past decade or so on grain distribution among modes have been completed. The basic objective of these studies was to alert the grain transportation industry to the powerful competitive forces developing in this sector, and this objective has been achieved. There has been a great demand for information on intermodal distribution of traffic, because no transportation agency having a large financial stake in its grain traffic can afford to overlook conditions attending this traffic.

Many of the new rates proposed by the railroads are titled "cost-oriented" rates, yet the relationships among some of these rates and apparent costs to the railroads of providing the services are not logically consistent. A preliminary test of the hypothesis that the rates are really oriented to the costs of competing carriers, rather than to the costs of the proponent railroads, indicated that the hypothesis merited further testing.

Grain

The grain marketing system, along with flour milling, is currently faced with problems of both expansion and contraction. Near depletion of whole grain stocks for export have left many terminal centers with relatively little business, while the Gulf Ports are constructing new facilities to handle the increased export flow. Terminals are moving closer to producers and flour mills are relocating closer to consumers. Milfeed users are also caught in this rapid shift in industry structure and location.

Research findings on the location and performance of the whole grain industry in the Northeast have been made available; a report prepared under contract by Oklahoma Agricultural Experiment Station on the structure, location, and grain flow in the Southwest is nearing completion; an analysis was completed for the 11 Western States and research is currently underway in the South; and Texas Transportation Institute, under contract with the Department and in cooperation with the 12-State Governors' Committee, is analyzing grain, oilseed, and flour transportation charges by motor carrier and barge in the North Central and Midwestern States.

C. Products and Services

Work was initiated in the past year to assess the impact of synthetics on markets for grain and fiber products and evaluate technical and economic factors that have contributed to market losses in certain areas. Data are being compiled on end-uses, prices, trends, market requirements, and competitive conditions in several important markets where these products are threatened by synthetic materials. Information developed will help identify and evaluate ways of strengthening the competitive position of agricultural products in industrial uses.

One market receiving special attention is that of starch from cereal grains in textile processing uses. Starch's competitive loss to synthetic chemicals over the last decade probably was most acute in fabric printing and somewhat less marked in textile finishing processes. The trend is toward such finishes as water-proofing, mildew-proofing, fire-proofing, wash-and-wear, and durable-press areas in which synthetic resins have gained prominence. Starch apparently has a solid economic advantage in textile warp sizing--the major use of starch in textile processing. Starch's use in this application will likely grow, offsetting losses in other areas, but its future as a warp size is by no means assured.

Declining per capita consumption of wheat and flour products in recent years has spurred interest in product innovations in this area. A market study on frozen bakery products and doughs indicated the number of firms producing these products increased from 8 in 1961 to 26 in 1965. Prices for white frozen dough averaged 15 to 20 percent lower throughout the United States than for the baker's regular white pan bread. The outlook is for continued steady growth in the production of frozen doughs.

D. Merchandising and Promotion

Research was expanded to develop and test a system using improved techniques to guide feed manufacturer management in making routing procurement and production decisions. The three major components of the system are: (1) forecasting sub-systems (production, sales, and prices); (2) inventory policy evaluation sub-system; and (3) allocation sub-system. This system has been tested and found feasible by a major midwestern feed manufacturer. Techniques and systems developed appear applicable to other agricultural firms.

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CONSUMER PREFERENCE AND QUALITY DISCRIMINATION --
HOUSEHOLD AND INDUSTRIAL
Standards and Research Division, SRS

Problem. Domestic consumption of agricultural commodities depends on the behavior of some 190 million consumers. But, in our complex marketing economy, it has become almost impossible for consumers to discuss their preferences, opinions, and dissatisfactions with producers and marketers. Knowledge of consumer reactions to agricultural products is becoming increasingly important because we are in a period of rapid change: There is a growing challenge to farm products and farm income from a wide variety of competitive products of nonagricultural origin; there is a proliferation of mixtures, forms, processes and other innovations affecting farm products; and there is increasing awareness that mistakes in developing, producing, and marketing farm products are costly not only to the farmer but to processors and handlers as well. An understanding of consumer reactions and the reasons behind them is essential to planning improvements in the production, marketing, and processing of agricultural products, developing educational programs, setting or revising grades or standards, evaluating new products developed by the Department's Utilization Laboratories, and identifying areas on which technical research should be focused to provide farm products in the forms and with the characteristics that will increase consumer acceptance and more closely satisfy consumer demand.

USDA AND COOPERATIVE PROGRAM

The Special Surveys Branch provides the consumer, in a scientific and unbiased manner, with an opportunity to say what he or she thinks about agricultural products by conducting applied research among representative samples of household, industrial, or institutional consumers and potential consumers. Such research may determine preferences, opinions, buying practices, and use habits with respect to various agricultural commodities; the role of competitive products; acceptance of new or improved agricultural products, consumers' ability to discriminate among products with varying attributes, and the preferences associated with specific forms. These studies of the opinions, preferences, knowledge and habits of consumers which affect their purchase and use of farm products provide a line of communication from consumers back to those concerned with production and marketing, and are complementary to the marketing and economic research of the Economic Research Service and the Consumer and Marketing Service as well as to utilization research of the Agricultural Research Service.

In addition to conducting studies of consumer preference and discrimination, the Branch also provides consultants and conducts special studies, upon request, for other agencies in the USDA or within the Federal Government, when survey methods can be usefully applied to the evaluation of programs, services, or regulatory procedures of interest to the requesting agencies.

The research is carried out in cooperation with other USDA or federal agencies, state departments of agriculture, experiment stations, land-grant colleges, and agricultural producer, processor, and distributor groups. Closely supervised contracts with private research firms are used for nationwide surveys; studies in selected areas are sometimes conducted by the Washington staff with the assistance of locally recruited personnel.

The Branch maintains all of its research scientists, who are trained in social psychology or other social sciences, in Washington, D. C., which is headquarters for all the research whether it is conducted under contract or directly by the Branch. The Federal scientific effort devoted to research on consumer preference for cereal products during the past year totaled 0.9 scientific man-years.

REPORT OF PROGRESS FOR USDA AND COOPERATIVE PROGRAMS

Consumer Preference

Potatoes, Rice and Wheat. The field work and coding of data on a nationwide study collecting information from homemakers on their use of and opinions about selected potato, rice, and wheat products have been completed, and preliminary runs of data are currently in process. A final report of the findings of this study will be published during the latter half of 1967.

PUBLICATIONS -- USDA AND COOPERATIVE PROGRAMS

None

IMPROVEMENT OF CROP ESTIMATING PROCEDURES
Standards and Research Division, SRS

Problem. Modern technology has been responsible for the development of new and improved methods of data collection and estimation. Sample surveys based on probability sampling are more expensive to conduct than the traditional self-selecting mailed survey but they do provide unbiased estimates and their precision can be measured from the sample data itself. The critical need for precision estimates for the agricultural economy makes it imperative that modern statistical theory and methods be developed and incorporated into the collection and analyzation of agricultural statistics. Many new techniques have been developed and introduced into the estimating procedures. However, due to the necessary demand for more and better statistics there is an urgent need for continued research and study to devise more efficient sample survey methods to insure continued improvement in the quantity and quality of SRS statistics.

USDA AND COOPERATIVE PROGRAM

The Department of Agriculture conducts a program of applied research designed to strengthen and improve the methodology used in collecting agricultural statistics. The principal disciplines involved are mathematics, statistics and probability but other disciplines relating to a particular subject or field are employed as required. Examples of these subjects are plant physiology, psychology, cartography and photogrammetry. The current program consists of 4.0 scientific man-years working on methods for forecasting and estimating the yields of important crops including grains and forages. Work under this program is done in Washington, D. C., and in SRS field offices located in the States concerned.

REPORT OF PROGRESS FOR USDA AND COOPERATIVE PROGRAMS

A. Wheat. The 1966 wheat objective yield program was increased to 2,410 samples in 17 States with the addition of 170 winter and 20 spring wheat samples. The sample allocation to States changed slightly from the preceding year based on a compromise between optimum allocation and the necessity to achieve a reasonable standard error for each State.

No weekly development studies for the entire growing season were conducted during 1965 as efforts were directed at analyzing data collected for earlier years. One of the purposes of the weekly development work was to provide information for determining the optimum size and shape of sample plots. Analysis of data collected does not support changing current size and shape of plot from the standpoint of variance only. However, this study is not completed as cost factors have not been completely analyzed.

Analysis of previous data for monthly samples in preparation of forecasting models for the 1966 crop, was done by a comprehensive computer edit and analysis programs. These programs greatly expanded the combinations of variables that could be tested and the different relationships tested for variables used in forecasting the components of yield.

Some work has been completed and study is continuing on the effective use of stratification to increase the precision of early season forecast and preharvest estimates of yield. Irrigated vs. non-irrigated growing areas, environment factors, geographic location, and variety are the stratification criteria under consideration.

Last year studies were undertaken to (1) improve early season counts and measurements for characteristics used in forecasting models for spring wheat, (2) determine the amount of gleanings on the ground prior to harvest and (3) consider the feasibility of counting immature grain by oven drying sample heads prior to threshing.

Plant counts were made in spring wheat fields in North Dakota, Idaho, and Washington on three weekly visits that covered the time period from plant emergence to the time when the stalks reached the flag or early boot stage of maturity. Prior studies indicated that plant count might be a more stable characteristic than stalk count during this part of the growth cycle due to rapid stooling. An analysis of this study indicated that plant counts were not consistently better than stalk count for measuring the number of potential heads to develop.

Preharvest gleanings were made for regular sample units in Montana and South Dakota. Enumerators were instructed to pick up heads, part-heads, and grain inside the sample unit on the final preharvest visit. The amounts gleaned although small were significant and during 1966 an adjustment was made to account for this difference in determining preharvest net yields.

At present, a ten head subsample from one row outside of one unit of the sample is being used to obtain information to forecast grain weight per head for the sample. The data obtained from this small subsample is highly variable in some instances. Grain count per head is one of the characteristics measured from this subsample. The Oklahoma Regional Laboratory conducted a study on counting grains from immature heads electronically. Heads for the study come from a randomly selected subsample of the regular monthly samples in North Dakota and Washington. All heads from an entire row length outside of one unit were clipped when the maturity stage of a sample was classified as being milk or soft dough. In the laboratory these heads were oven dried prior to threshing. The number of grains that could not be threshed from the heads was significant as compared with the total grain count from heads. Further research to improve counting procedures should be initiated.

During 1966, a plant development study was conducted in two States (Oklahoma and Oregon.) The purpose of this study is to count and measure plant characteristics associated with the development of heads and to collect information relating to the influence of environmental conditions on this factor. Four fields were observed in each State and two samples were located within each field. Weekly visits were made to each field from May 1 until harvest.

B. Pasture grasses. Studies aimed at measuring the amount of forage produced and the amount consumed by livestock were conducted under a cooperative agreement with Iowa State University were continued in 1965. Work done in the earlier years had been concerned with determining optimum procedures for estimating growth and intensity. This was done through experimental designs which allowed analysis by cage types (mobile or stationary), plot shapes, time period of observation (one, two, or four weeks), as well as appropriate field techniques and cost structures. The work prior to 1965 was done on University farms and indicated plot variation within pastures and variation among farms were both important sources of variation in seasonal forage production and grazing.

In the summer of 1965, a larger scale study was initiated. This study covered 16 farms in 3 Iowa Crop Reporting Districts. Three plots per farm pasture were randomly located. Each plot consisted of a 3' X 3' cage surrounded by four outside plots. The outside plots were used to determine the amount of grazing and forage available at the start of the study. The caged area determine the production during the time period.

The 1965 study clearly demonstrated the existence of a between farm component of variation in addition to an intra-farm component. It also indicated less variability in grazing estimates than in growth estimates. The 1965 study, as well as earlier, studies, showed that the caged-plot technique using mobile cages provides a feasibly operational method for measuring pasture yields.

PUBLICATIONS -- USDA AND COOPERATIVE PROGRAMS

None

ECONOMIC AND STATISTICAL ANALYSIS
Economics and Statistical Analysis Division, ERS

Problem. Frequent accurate appraisals of the economic prospects for important agricultural commodities are necessary if farmers are to plan and carry out their production and marketing activities in an efficient and profitable way. The typical farmer cannot afford to collect and analyze all the statistical and economic information necessary for making sound production and marketing decisions. Such information is provided through a flow of current outlook information; the development of longer range projections of the economic prospects for the principal agricultural commodities; and analyses of the economic implications of existing and proposed programs affecting major farm commodities.

Producers, processors, distributors, and consumers need information based on accurate quantitative knowledge of the interrelationships among prices, production and consumption of farm products, and other factors. Similarly, Congress and the administrators of farm programs need such economic information to evaluate existing and alternative programs or policies in terms of their probable impact on production, consumption, and prices at both the farm and retail levels. The research program in this area provides the information for strengthening outlook and situation work, and for appraising alternative policies for agricultural products.

USDA AND COOPERATIVE PROGRAM

The Department has a continuing program of basic research concerning the factors affecting prices, supply, and consumption of principal agricultural commodities and the analyses of the situation and outlook for selected commodities. The Federal scientific man-years involved for grain are 3.5 annually, of which 1.0 are devoted to work on the supply and demand for grains and 2.5 to work on the grain situation and outlook. The program is carried on in Washington, D. C.

PROGRAM OF STATE EXPERIMENT STATIONS

For the most part the States depend heavily on the USDA for across-the-board commodity situation and outlook research. However, the State extension staff members supplement and adapt such research information to meet the commodity situation of their States. The total direct research effort at state Experiment Stations in the situation and outlook area is small--probably no more than 2 to 3 scientific man-years. While not designed as outlook research, much of the research conducted by the experiment stations contributes to improved understanding of price-making forces, which in turn improves market situation analysis and price forecasting.

Many of the States carry on supply, demand, and price analyses for the products of their State. Much of the research is commodity oriented, though some projects are of a highly mathematical and theoretical nature aimed at improving price analysis methodology.

PROGRESS -- USDA AND COOPERATIVE PROGRAMS

A. Situation and Outlook Analysis

1. Food Grains

During the past year considerable resources were employed in the analysis of alternative wheat and rice programs. With the emergency of food shortages in India, emphasis was placed on alternative food aid programs, including the substitution of other grains for wheat. The decline in wheat stocks, both in the U.S. and around the world, called for additional attention to world wheat prices, U.S. export pricing policy, and provisions of the 1967 Wheat Program. Considerable material was furnished the National Food Marketing Commission at their request.

The situation reports reflected expanded work on the feeding of wheat to livestock; the supply and demand situation in major wheat producing nations competing with the U.S. in world trade; increased trade by other wheat exporting countries with the communist nations and the effect on the U.S. situation; and the rye situation in the U.S. Formerly, rye was covered only intermittently in the Wheat Situation but in the last year it has been discussed in each issue.

2. Feed Grains

The Feed Grain Program continued to be given special attention in 1965-66. Much of the analysis has been done to aid in establishing a desirable production-utilization balance through use of a voluntary type acreage diversion program. Feed grain stocks have been materially reduced since the beginning of the program in 1961 when a record 85 million tons was on hand. Feed grain requirements have exceeded production in 4 out of the last 5 years, reducing the carryover in 1966 to about 47 million tons. With prospects for another decrease in carryover in the 1966-67 season, alternative programs are under study. Annual average prices received for feed grains have risen about 16 percent from the low of 1960-61. In the past 3 years, prices have been above the loan rates, resulting in much less grain going into the program and a marked reduction of Government stocks through CCC sales.

B. Demand, Supply and Price Analysis

1. Feed Grains and Other Feeds

A special study was made of major factors influencing high-protein feed prices. Much of the analysis was directed to soybean meal, but factors associated with prices of cottonseed meal, linseed meal, gluten feed, meat

meal and fish meal also were studied. The four factors that stood out as the most important in influencing prices of these high-protein feeds were: the quantity of soybean meal or total high-protein feeds fed, the index of prices received by farmers for livestock and livestock products, the number of high-protein consuming animal units, and production of commercially prepared feeds. The results of this analysis were presented in the April issue of the Feed Situation. Another special study was conducted on trends in supplies and prices of hay and roughage consuming livestock by major regions of the United States. Results were published in the August issue of the Feed Situation.

2. Food Grains

During the past year increased work has been carried out on the measurement of demand and price for wheat, rice and rye. With the loan program having diminished as a price-making force in the wheat markets, and with increased use of wheat for feed, new price estimating methods have been developed. These techniques take greater account of privately held stocks of wheat and the feed grain situation generally; they are more concerned with individual areas and classes of wheat than formerly was the case. A study was made for the State Department dealing with rice in Southeast Asia and the world rice outlook over the next five years.

PUBLICATIONS -- USDA AND COOPERATIVE PROGRAMS

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